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WILDFIRE DISEASE OF SOYBEANS[1]

WILLIAM B. ALLINGTON[2]

(Accepted for publication June 10, 1945)

INTRODUCTION AND REVIEW OF LITERATURE

An intensive soybean disease survey was made by the United States Regional Soybean Laboratory in the summer of 1943, covering many of the heavy producing areas. Of particular interest was a disease identified as wildfire, caused by *Pseudomonas tabaci* (Wolf and Foster) Stapp.

Three references in the literature report soybeans (*Soja max* Piper) susceptible to this parasite when artificially inoculated (1, 8, 10). In one instance attempts to re-isolate the organism and establish definite proof of pathogenicity were not reported (8). Valleau *et al.* (10) have reported ten successful passages of the organism in soybeans growing in the greenhouse. To the writer's knowledge, however, the disease has not been previously recognized to cause extensive damage to soybeans in the field. The total damage observed in 1943 and 1944 was not significant. However, in individual fields, damage was severe enough to make evident the potentialities of this disease on soybeans and to emphasize the need for careful study.

SYMPTOMS

Symptoms are so characteristic that this criterion alone is usually sufficient for identification of wildfire. The necrotic spots on the leaves are variable in size and are nearly always surrounded by a striking wide yellow halo (Fig. 1). At times the halo, as on tobacco, may be rather indistinct. This condition is thought to be the result of rapid invasion by the bacteria to cause a breakdown of the tissue before the chlorophyll-destroying exotoxin has had time to destroy the chlorophyll. In such instances the necrotic spot is dark brown to black as contrasted to the usual light brown. The lesions, in suitable damp weather, tend to enlarge and coalesce, and eventually they involve large areas of the leaf. Defoliation, starting on the lower leaves, is common and under certain conditions may progress until few leaves remain. So far as is known the infection on soybean is confined to the leaves.

The infection in most fields is usually limited to definite areas, which may be a few feet in diameter or several acres. Few fields have been observed to be uniformly infected throughout. This is thought to be partly due to variation in environmental factors such as soil fertility and drainage.

[1] A publication by the U. S. Regional Soybean Laboratory, a cooperative organization participated in by the Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration; and the Agricultural Experiment Stations of Alabama, Arkansas, Florida, Georgia, Illinois, Indiana, Iowa, Kansas, Louisiana, Michigan, Minnesota, Mississippi, Missouri, Nebraska, North Carolina, North Dakota, Ohio, Oklahoma, South Carolina, South Dakota, Tennessee, Texas, Virginia, and Wisconsin.

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OCCURRENCE AND DISTRIBUTION

Wildfire was observed in all the major soybean-producing areas, except possibly North Carolina, in 1943 or 1944. The first specimen to come to the writer's attention was collected in northern Mississippi in July, 1943. In early August, 1943, severe infections were found in Arkansas, Tennessee, Alabama, and Georgia. In late August and early September of the same year, infection was observed in Iowa, Illinois, Indiana, and Ohio. In 1944, the disease was reported on soybeans from most of the North and South

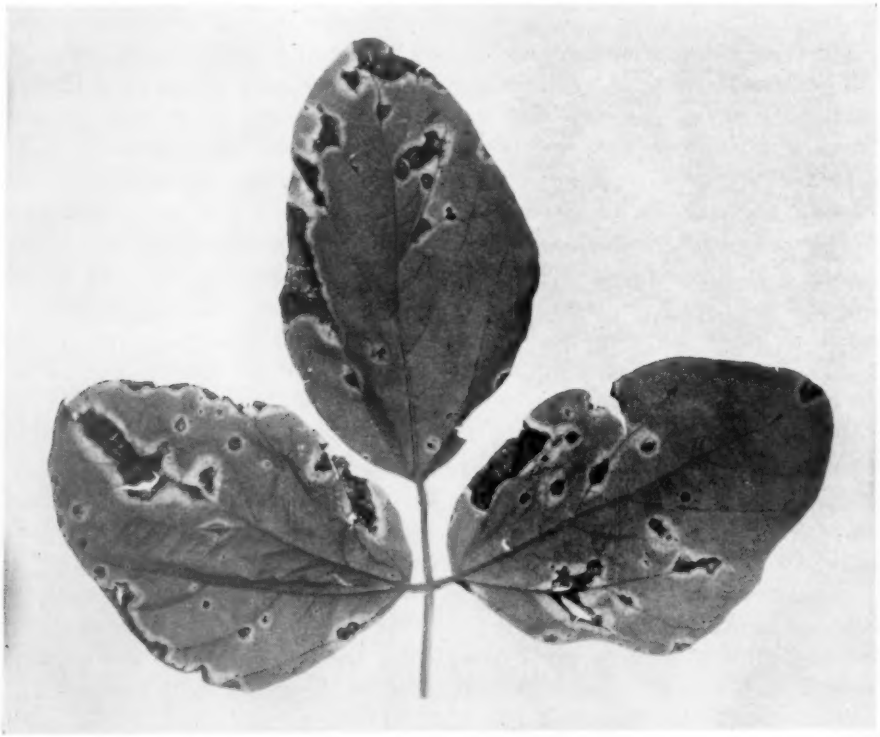

FIG. 1. Typical wildfire on soybean from natural infection in the field. The striking yellow halo around the necrotic spots is the chief distinguishing feature. Note coalescence of necrotic areas.

Central States. Extensive infection in 1943 and 1944 was rare before August, but it is not known whether wildfire is typically a late season disease on soybeans.

The most extensive infection observed to date over a wide area was in central Iowa in August, 1944. Wildfire is not known definitely to have occurred on soybeans before 1943, but it probably has been present in isolated areas for several years. Definite increase in the disease on soybeans over the country has been observed in the last two years. Should this continue, it may well be a menace to the soybean crop.

THE CAUSAL ORGANISM

Extensive tests have been made with the soybean wildfire organism to compare it with the tobacco wildfire bacterium. Studies of their morphological, physiological, serological, and pathological characteristics are here reported.

Morphology and Physiology.—Six isolates of the soybean wildfire organism were compared in pure culture with two isolates of *Pseudomonas tabaci* (Wolf and Foster) Stapp obtained from other laboratories. The cultural differences within each group were as great as the differences between the groups.

The soybean wildfire organism is a white, Gram-negative rod, $0.86 \times 3.0\ \mu$, motile by 1–5 polar and bipolar flagella. Spores are absent. The colony type is indistinguishable from *Pseudomonas tabaci* on potato-dextrose agar at pH 7.0. Spontaneous dissociation into rough and smooth colonies was frequent on the various culture media.

The soybean wildfire organism liquefied gelatin readily. Litmus milk was turned blue in 4 days, the litmus being eventually reduced with coagulation. Hydrogen sulphide was not produced in peptone broth. Indole was not produced in broth containing tryptophane (Gore method). Starch was not hydrolyzed. Green fluorescence was commonly observed on a variety of liquid media.

No definite differences were found between the soybean isolates and the tobacco isolates in their ability to utilize the following carbon and nitrogen sources: lactose, levulose, inulin, mannite, maltose, glycerol, l-arabinose, l-xylose, d-galactose, dextrose, sucrose, ammonium sulphate, potassium nitrate, potassium nitrite, l-cystine, glycine, succinimide, acetamide, and urea. The results were in substantial agreement with those reported by Braun (2). Slight variation was observed in the ability of the isolates to utilize these compounds. However, the difference between the soybean isolates and the tobacco isolates was not greater than the differences within each group.

Serology.—The agglutination and agglutinin absorption tests were used to further test the relationship of the soybean and tobacco wildfire organisms. The agglutinin absorption test is considered by many to be the most accurate test for the identity of organisms. However, it may be preferable to use these tests in conjunction with the others reported, and to draw conclusions in regard to relationships from the general results produced. Bacteria are known to undergo changes, in pure culture at least, which can influence the investigator to such an extent that he may be led to create new species or varieties or eliminate species and varieties unwisely, unless he has sufficient information of various types upon which to base his judgment.

The serological relationships of four soybean isolates and two tobacco isolates were compared. Isolates S1, S2, and S6 originated from diseased soybeans from central Illinois, while S7 was isolated from diseased soybeans from Alabama. Tobacco isolates T10 and T11 were obtained from the Divi-

sion of Tobacco Investigations, U. S. Department of Agriculture. Cultures were from single colonies but were not single-celled. Their pathogenicity was established and checked periodically during the experiment and they were always highly pathogenic to both soybeans and tobacco. Only the "smooth" forms of the cultures were used for injections.

Rabbits were used for the production of the immune sera. Before the first injections the normal sera from these animals were tested and found to be void of normal agglutinins for the organisms studied. The bacterial cultures used for the immunization of the rabbits and the serological tests were grown for 24 hours on potato-glucose agar. Suspensions of the bacteria for injection into the animals were made in 0.85 per cent NaCl solution and filtered through sterile filter paper. The turbidity was standardized to conceal a 2-mm. loop made of No. 26 B. & S. gauge wire at a depth of 12 mm.

TABLE 1.—*Results of agglutination tests with sera from rabbits injected with 4 isolates of soybean wildfire bacteria and two isolates of tobacco wildfire bacteria, Pseudomonas tabaci (Wolf & Foster) Stapp*

Antisera[a]	Maximum dilution at which agglutination occurred					
	Isolate					
	S1	S2	S6	S7	T10	T11
S1	1280	1280	2560	2560	2560	1280
S2	1280	1280	2560	2560	1280	1280
S6	2560	1280	1280	2560	640	640
S7	2560	1280	1280	2560	640	1280
T10	1280	1280	1280	1280	1280	1280
T11	2560	2560	2560	2560	2560	1280
Normal serum	0	0	0	0	0	0

[a] Antisera S1, S2, S6, S7 obtained from rabbits inoculated with soybean wildfire isolates. Antisera T10 and T11 from rabbits inoculated with two tobacco wildfire isolates. Homologous antisera and isolates bear the same numbers.

The rabbits received 7 intravenous injections, at intervals of 4 to 5 days. The amount of each injection was, respectively: 0.5 cc., 1 cc., 1.5 cc., 2.0 cc., 2.5 cc., 3.0 cc., and 4.0 cc. Ten days after the last injection, the rabbits were bled from the ear vein.

The results of the macroscopic agglutination tests are in table 1. Although the titre of the sera are slightly low, the data indicate the close relationship between the isolates from soybean and those from tobacco, and one cannot differentiate them by this test.

The agglutinin absorption test was made with the same antisera as the agglutination tests. The minimum absorbing dose of antigen was determined for each serum with its homologous organism. Bacteria were suspended in a phenolized saline solution (0.5 per cent phenol) which was adjusted to different densities varying from 3.0 to 8.0 on the McFarland nephelometer scale. One part of nondiluted antiserum was added to 39 parts of each of the suspensions, giving a serum dilution of 1–40 in each

case. The tubes were incubated at 37° C. for 2 hours, centrifuged, and the clear supernatant liquid removed to clean, sterile, centrifuge tubes. Bacteria from the packed centrifuge sediment of a broth culture were added to produce again the original turbidity in each tube. The tubes were again incubated at 37° C. for 2 hours, centrifuged, the clear supernatant liquid removed and reabsorbed with bacteria. After a subsequent 2-hr. incubation at 37° C. they were placed in the refrigerator over night. The following morning, they were centrifuged and the supernatant was used for agglutination tests with the homologous organisms, with antiserum dilutions through

TABLE 2.—*Results of agglutinin absorption tests with cultures of two soybean wildfire isolates and cultures of two tobacco wildfire isolates*

Antisera[a]	Absorbing antigen	Maximum dilution at which agglutination occurred			
		Isolate			
		S6	S7	T10	T11
S6	S6	.....	.....	.....	.....
	S7	.....	.....	.....	.....
	T10	.....	.....	.....	.....
	T11	.....	.....	.....	.....
S7	S6	.....	.....	.....	.....
	S7	.....	.....	.....	.....
	T10	.....	.....	.....	.....
	T11	.....	.....	.....	.....
T10	S6	.....	.....	.....	.....
	S7	.....	.....	.....	.....
	T10	.....	160	.....	.....
	T11	.....	.....	.....	.....
T11	S6	.....	.....	320	160
	S7	.....	320	80	.....
	T10	.....	.....	.....	.....
	T11	.....	.....	.....	.....

[a] Antisera S6 and S7 obtained from rabbits inoculated with soybean wildfire isolates. Sera T10 and T11 from rabbits inoculated with two tobacco wildfire isolates. Homologous antisera and isolates bear the same numbers.

640. The lowest turbidity to have completely absorbed the agglutinins was considered the minimum absorbing dose for that serum. By using suspensions about four times as heavy as the minimum absorbing dose, portions of each antiserum were absorbed with one of the six cultures. Each absorbed antiserum then was tested for agglutinins for all six isolates. The results of the tests are in table 2.

In most cases reciprocal agglutinin absorption occurred. It is believed that the five positive tests in table 2 are due to inherent variations in the method and are of no consequence, especially since there is no consistency. If these results do indicate differences in the organisms studied, one can safely say that the difference between the two tobacco isolates is as great as between the soybean and tobacco isolates.

Pathogenicity.—In view of the recent work on the relationship of watersoaking of tissue to infection by bacteria, caution should be exercised in the interpretation of data in regard to pathogenicity. This subject will be discussed later in the paper. The soybean wildfire isolates were introduced into watersoaked leaf tissue of tobacco or soybean in order to obtain infection. Practically no infection results with these or with the tobacco isolates without some degree of watersoaking. Figure 2 shows infection on tobacco with the soybean and tobacco isolates. After the development of typical wildfire symptoms on soybean and tobacco plants, portions of the infected tissue were macerated, diluted, and plated out on potato-dextrose agar at pH 7.0. The organism was readily reisolated. Subsequent passages and the number of bacteria obtained from the lesions left no doubt as to the causal agent.

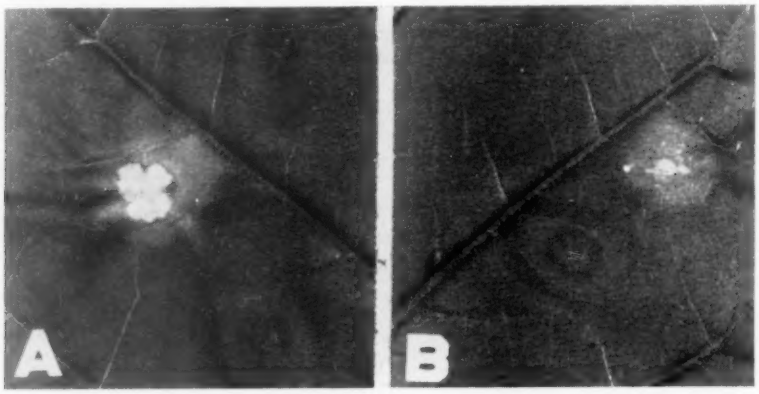

FIG. 2. A. Wildfire lesion on tobacco caused by an isolate from soybean. B. Wildfire lesion on tobacco caused by an isolate from tobacco. Tobacco was artificially inoculated by atomizing. Fundamentally, no differences existed between the isolates.

The virulence of the wildfire isolates on tobacco has been reported to vary greatly, and the soybean isolates were no exception in this respect. The ability of this parasite to kill tissue and to produce the powerful chlorophyll destroying exotoxin so characteristic of the organism varied extensively with the different isolates.

The exotoxin is produced abundantly in pure culture, and by heating as well as by filtration the organism was separated from it. The exotoxin was tested several times on living plants in the greenhouse and found to be active when introduced into leaves by the needle prick method.

Extensive inoculation tests in the greenhouse and field for two years have failed to disclose any difference in symptoms produced by the soybean and tobacco wildfire isolates.

THE INFECTION PROCESS

A new and not too generally recognized aspect of infection by leaf-spot bacteria, i.e., the role of watersoaked tissues, has been studied and reported

on by a relatively few investigators (3, 6, 10). The writer has found watersoaking of great consequence in the penetration of soybean leaves by *Pseudomonas tabaci.*

Soybean leaves become watersoaked in the field either by a beating rain or by internal physiological phenomena. Often this watersoaking is of limited extent and of short duration, and can easily pass unnoticed. Diachun *et al.* (5) have described the manner in which liquids, containing chemicals, ink, etc., penetrate watersoaked tobacco leaves. Approximately the same experiment was performed with soybeans. Three methods of artificially watersoaking soybean leaves were employed in this study with the aim of simulating physiologic and storm watersoaking. Obviously natural conditions cannot be duplicated exactly but it is hoped that the methods, materials, and results reported here will suffice to demonstrate the writer's views on the infection phenomena as they occur in nature.

Two methods have been employed to watersoak leaf tissue experimentally without beating the water against the epidermis as occurs in heavy rains. One is the hypodermic needle method (Fig. 4, C). Water can readily be injected into the intercellular spaces by inserting the needle near a vein on the lower leaf surface. The other method is the insertion of the cut stem of the plant into a water-tight connection and its subjection to high water pressure as described by Johnson (6). Both methods produce a physiologic watersoaking similar to the type occurring in the field in the absence of beating rain.

Watersoaked tissue will absorb a liquid from the surface of the leaf very readily when a junction is established between the intercellular and the external liquid (Fig. 3). Penetration and dispersion within the watersoaked area of the leaf is very rapid, usually requiring only a few seconds. The forces operating to pull the liquid into the leaf are unknown but appear to be related to capillarity. Liquid junctions, resulting in bacterial penetration apparently are not so common in this type of watersoaking as in the storm type. Without mechanical injury, drops of bacterial suspension or other liquids may stand indefinitely on a physiologically watersoaked area without penetration occurring (Fig. 4, A and B). If the water by the application of internal pressure has been forced out through the stomata or old wounds in the leaf then penetration is immediate. This rarely is observed in nature. On healthy soybeans in the field, physiological watersoaking is usually confined to the base of the leaflet at the point of attachment. It may be observed early in the morning, during the growing season, especially if heavy dew is present. However, in severe epidemics of wildfire, only a few of the leaves may have characteristic infections at the base where this watersoaking has occurred. It is doubtful, therefore, if this type of watersoaking is of much consequence as far as initial infection of soybeans is concerned. Johnson (7), working with tobacco, has concluded that this type of watersoaking is very important in initial infection, and that prolonged periods of watersoaking are essential for the production of large lesions.

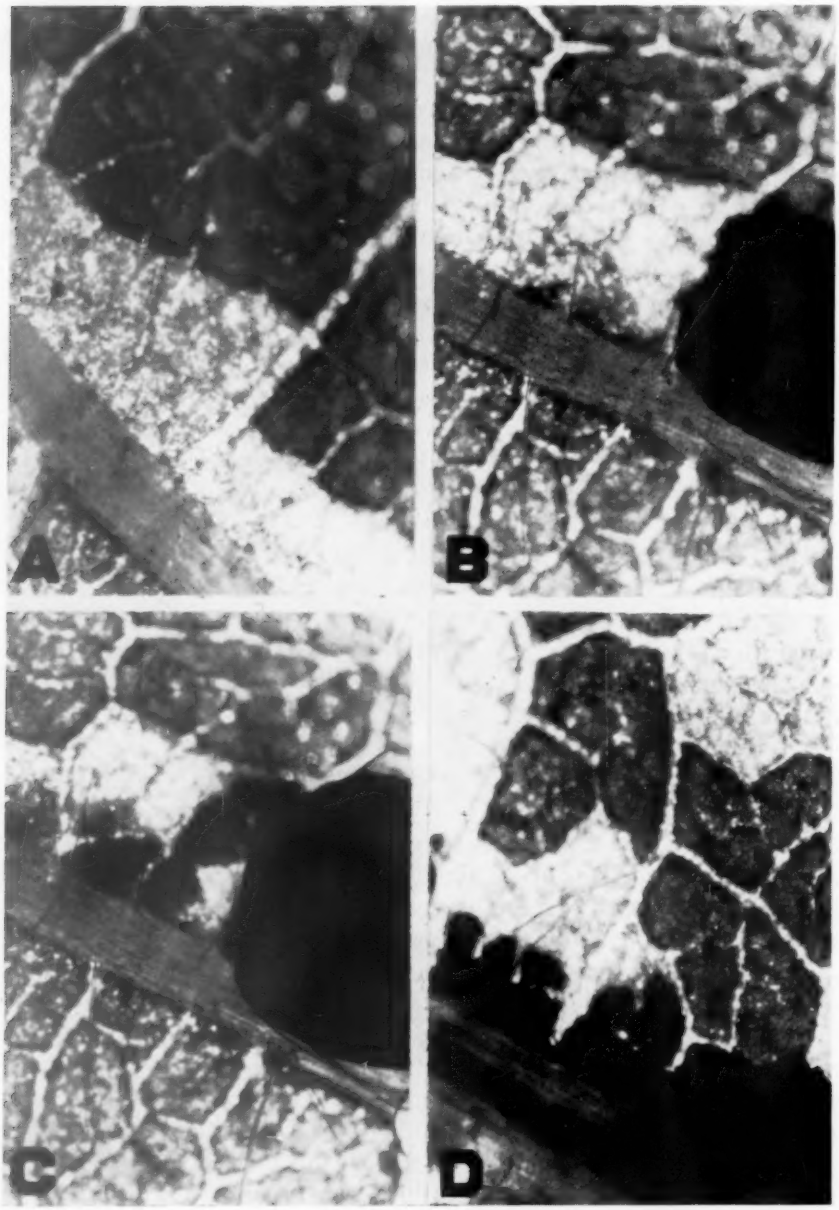

FIG. 3. Photomicrographs showing the progress of India ink in soybean leaf tissue artificially watersoaked by the hypodermic needle method (Fig. 4, C). Photographed with transmitted light, so that the translucent watersoaked tissue appears light as contrasted to the normal green tissue. A. Watersoaked tissue along a large vein. B. India ink has been placed over an unwounded portion of the watersoaked area. C. After wounding, the ink penetrates the leaf tissue very rapidly. D. Further penetration of ink. The process of penetration, as illustrated in C and D, may take place in less than 10 seconds. (×10.)

Beating rains are observed to be closely connected with wildfire epidemics on soybeans. Under these circumstances, the liquid junction is automatically made between intercellular and surface water when the water is driven into the leaf. Figure 4, A and B, demonstrates how the two types of watersoaking respond when India ink is placed over the watersoaked area when care is taken not to puncture the epidermis with sharp instruments. When watersoaking occurs, there may or may not be mechanical injury to the tissues. Observations have indicated that the condition of the plant is an extremely important factor in regard to the amount of infection. For

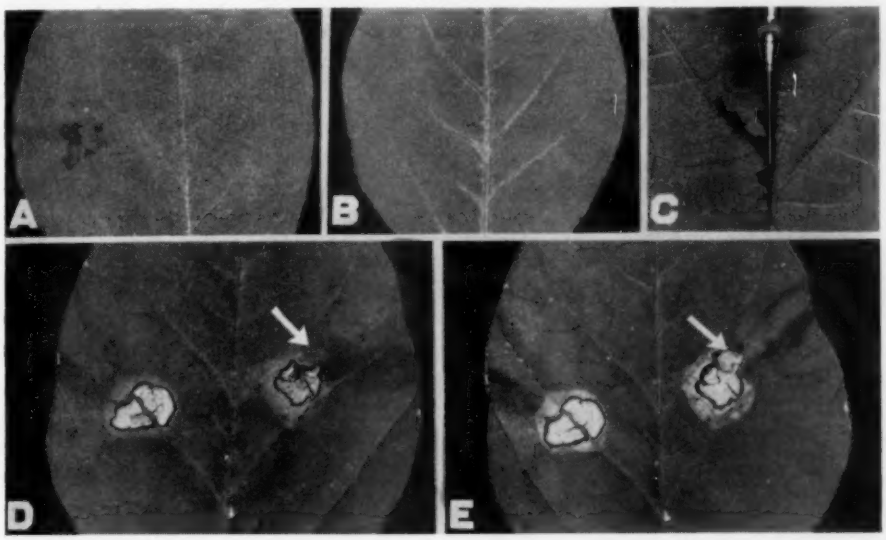

FIG. 4. Soybean leaves illustrating various relationships of watersoaking to infection and development of wildfire. A. A small area was watersoaked by atomizing water against the lower surface. India ink was then placed over the watersoaked tissue for five minutes as well as over the non-watersoaked tissue on the opposite side of the leaf, then gently washed off. Penetration of ink can only be observed where watersoaking existed. B. Leaf previously watersoaked by water pressure from water line through cut stem. India ink was placed over the watersoaked areas on this leaf, as in A, for five minutes. No penetration can be observed. C. Method of watersoaking leaf tissues by use of the hypodermic needle. D and E. Wildfire lesions on the same leaf. E was photographed five days later than D. Inoculation was made by atomizing the bacterial suspension against the lower surface where the infected areas developed. One week after inoculation, the area indicated by the arrow in D was watersoaked as illustrated, by atomizing sterile water. Five days later, as shown in E, the lesion occupied part of this previously watersoaked area (arrow in E). Watersoaking persisted thirty minutes. Note similarity of the lesion on the left in D and E.

example, the condition of the stomata is important as shown by Diachun (4) with tobacco. One year's field trials indicate that soybeans sprayed with bacterial suspensions with a power sprayer are apt to have greater infection if sprayed during the bright sunshine than if sprayed just before daylight or at dusk in the evening. In this connection, it is interesting that at certain times, for example at night, when the stomata on soybeans are closed, storm watersoaking is difficult to produce artificially, yet it is easy in the

daytime. It would appear therefore that the water is driven in through the stomata, and it is only logical to conclude that bacteria penetrate mainly by the same route. On the other hand, physiologic watersoaking occurs on soybeans primarily at night or on extremely dark days, and extensive penetration through stomata in this case would be difficult to explain.

The spread of bacteria through leaf tissue is greatly enhanced by watersoaking, as one might conclude from the behavior of ink in watersoaked tissue (Fig. 4, A). This is evident as shown in figure 4, D and E, which illustrates the result of an experiment repeated many times. A leaf was inoculated with a very dilute suspension of wildfire bacteria from a culture 24 hours old. The suspension under considerable pressure was atomized over a small area, thereby watersoaking the tissue in that area and assuring good penetration. One week after inoculation typical wildfire lesions appeared on the leaf, as in figure 4, D. At this time normal tissue adjacent to one of the lesions was watersoaked by atomizing sterile water against the leaf at high pressure. This was done in sunlight and the plant was never removed from the greenhouse bench. Watersoaking persisted for less than 30 minutes. The leaf was rephotographed after 5 days and appeared as in figure 4, E. The bacteria had penetrated the watersoaked tissue in a very short time and the result was an increase in the size of the necrotic spot with a definite zonation at the margin of the old lesion. The lesion that had not received a second watersoaking remained substantially the same, except that slight intensification and growth of the halo was evident after 5 days. Physiologic watersoaking has been observed to operate in a manner very similar to this. Likewise, water from dew absorbed at the margin of static lesions, may cause a very narrow ring of watersoaking and enhance the subsequent spread of bacteria within the normal tissue; the result is reactivation and growth within the lesion, and at times concentric rings are formed.

Penetration and dispersion of the bacteria in watersoaked leaf tissue is surprisingly rapid. Judging from the experimental results obtained, prolonged watersoaking is not essential for the establishment of infection after penetration.

An attempt was made to check directly upon the number of bacteria present in leaf tissue held under various conditions. Soybean leaves were completely watersoaked by directing an atomizer, with sterile water, on the lower surface of the leaves. Leaves of comparable age and position on the plant were used as non-watersoaked checks. A very dilute suspension of *Pseudomonas tabaci* from a 24-hour potato-dextrose-agar culture was made in a sterile 500-cc. beaker. The watersoaked and non-watersoaked leaves were immersed together in the suspension for 1 minute after which they were exposed to the air for 3 minutes. They were then washed 8–10 times in large beakers of sterile distilled water. Half of each watersoaked leaf was then placed between moist blotters in a moist chamber, the other half was dried in the air for 20–25 minutes, until the watersoaking disappeared. At this time, the non-watersoaked leaf and the half leaf which had

recovered from watersoaking were placed beside the continuously watersoaked leaf between the blotters in a moist chamber. The leaves were subjected to identical conditions except for the watersoaking. After one hour, 10 discs cut at random from each leaf with a sterile cork borer of 6 mm. diameter were completely macerated in 10 cc. sterile water, and adequate dilution was made to facilitate counting of colonies on plates containing 1 cc. of suspension.

The results as shown in table 3 illustrate several facts concerning the role of watersoaking. Obviously penetration was greatly dependent upon watersoaking. The rate of multiplication of the bacteria was greatest within the continuously watersoaked tissue. However, the tissue which was

TABLE 3.—*The effect of watersoaking on penetration and the subsequent growth by wildfire bacteria*

Exp. No.	Average number bacteria per disc in 10 leaf discs from non-watersoaked leaves		Average number bacteria per disc in 10 leaf discs from watersoaked leaves		
				After 24 hours	
	After 1 hr.	After 24 hrs.	After 1 hr.	Watersoaked 1 hr.	Watersoaked 24 hrs.
1	3.2	0.0	19.6	29700.0	13900.0
2	1.0	20.5	120.6	5781.0	31700.0
3	0.0	0.2	12.0	200.0	950.0
4	50.0	58.0	208.0	42476.0	56175.0
5	5.1	23.3	579.0	180.0	2488.0
6	2.5	0.9	16.5	120.0	5750.0
7	0.0	0.0	27.8	2045.0	4844.0
Ave.	8.8	14.7	140.5	11643.1	16543.8

watersoaked only during the time of inoculation also supported rapid multiplication. There is so much variability in the results that it is questionable whether this difference with duration of watersoaking is significant. During the time the leaves were recovering from watersoaking, occasionally certain leaves lost more water than others, as judged by appearance. This may be a factor in the variability encountered. It appears that watersoaked or previously watersoaked tissue might be a more favorable place for bacterial growth than tissue that never has been watersoaked. The general averages would indicate approximately one generation in 24 hours in the tissue which never had been watersoaked, as contrasted to between 7 and 8 generations in the tissue previously watersoaked. Considerably more study is necessary to definitely establish this fact.

DISCUSSION

The common occurrence of wildfire on soybeans in the last two years raises many questions in regard to its origin and dissemination. Its occurrence on soybeans never has been reported to be connected with tobacco culture. Three fundamentally different explanations have been advanced to

account for the source of wildfire inoculum for tobacco each year. These explanations are that the inoculum comes primarily from the following: first, the previous year's infected refuse or seed; second, from the soil where the organism has been found to grow epiphytically upon many crop plant roots in addition to lying dormant in the infected refuse or seed (10); and third, from the soil where the organism exists primarily as the common soil saprophyte *Pseudomonas fluorescens,* mutating or changing suddenly in some manner to become parasitic and to possess all the properties of *Ps. tabaci* (9). With regard to soybeans the second explanation seems to be the most plausible with the exception of seed transmission. No infection on soybean seed or pods has been observed and no instance of suspected seed transmission has, as yet, been reported.

The incidence of wildfire in isolated spots in large soybean fields, hundreds of miles away from any tobacco growing areas, immediately implies some origin other than previous crops of tobacco. Perhaps the explanation advanced by Valleau *et al.* (10) for tobacco wildfire, that is, epiphytic growth on various plant roots, could apply to the disease on soybeans. The hypothesis concerning the possible relation to *Pseudomonas fluorescens* would explain its occurrence; however, it is considered unlikely that the soil saprophyte can readily change to a parasite as virulent as *Ps. tabaci.*

Until further research has been done no control measures can be recommended. The search for resistant varieties has progressed only one year and resistance has not yet been found.

The serological and pathological results obtained indicate that variability exists in the wildfire organism. However, since this is observed within the group of isolates from soybean and within the group from tobacco, it is reasonable to conclude that the two groups are not sufficiently different for specific or varietal differentiation.

The discovery of the invasion of watersoaked tissue by pathogenic bacteria (3) explained many phenomena in regard to bacterial leaf spots heretofore unaccounted for. A common misconception, however, has been that the tissues concerned must remain watersoaked for several hours. This is not the commonly observed fact in nature and has perhaps delayed the general acceptance of the importance of watersoaked tissue. The importance of physiologic watersoaking on large soybean plants in the field is questionable in so far as it concerns initial infection. On the other hand, the beating of rain on the leaves when the stomata are open, sometimes causes extensive watersoaking which may be followed by severe epidemics of wildfire.

SUMMARY

Wildfire, caused by *Pseudomonas tabaci,* is common on soybeans in most of the commercial soybean growing areas of the United States. The damage in isolated areas is severe. Morphologically, physiologically, serologically, and pathologically the organism isolated from soybeans does not differ from

isolates of *Ps. tabaci* from tobacco, and the two diseases are considered to be caused by the same organism. Watersoaking of soybean leaf tissue greatly enhances penetration of the leaves by *Ps. tabaci* and spread of the bacteria through the tissue. Physiologic watersoaking was not so effective as storm watersoaking in bringing about penetration. Prolonged watersoaking was not necessary for the growth or dispersion of bacteria within the tissues.

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LITERATURE CITED

1. BELTYUKOVA, K. I., and O. P. LEBEDEVA. On the specialization of *Phytomonas tabaca* Wolf and Foster on certain host plants. The A. I. Mikoyan Pan-Soviet Sci. Res. Inst. and Indian Tob. 2nd. Krasnodar Publ. 126. 17–34. 1936.
2. BRAUN, A. C. A comparative study of *Bacterium tabacum* Wolf and Foster and *Bacterium angulatum* Fromme and Murray. Phytopath. **27**: 283–304. 1937.
3. CLAYTON, E. E. Watersoaking of leaves in relation to development of the wildfire disease of tobacco. Jour. Agr. Res. [U. S.] **52**: 239–269. 1936.
4. DIACHUN, STEPHEN. Relation of stomata to infection of tobacco leaves by *Bacterium tabacum*. Phytopath. **30**: 268–272. 1940.
5. ———, W. D. VALLEAU, and E. M. JOHNSON. Invasion of watersoaked tobacco leaves by bacteria, solutions, and tobacco mosaic virus. Phytopath. **34**: 250–253. 1944.
6. JOHNSON, J. Relation of watersoaked tissues to infection by *Bacterium angulatum* and *Bacterium tabacum* and other organisms. Jour. Agr. Res. [U. S.] **55**: 599–618. 1937.
7. ———. Bacterial invasion through stomata. (Abstract) Phytopath. **34**: 11. 1944.
8. ———, C. M. SLAGG, and H. F. MURWIN. Host plants of *Bacterium tabacum*. Phytopath. **14**: 175–180. 1924.
9. REID, J. J., J. NAGHSKI, M. A. FARRELL, and D. E. HALEY. Occurrence and nature of the microorganism associated with wildfire. Pa. Agr. Expt. Sta. Bull. 422. 1942.
10. VALLEAU, W. D., E. M. JOHNSON, and S. DIACHUN. Angular leafspot and wildfire of tobacco. Ky. Agr. Exp. Stat. Bull. 454. 1943.

LEAFHOPPER OVIPOSITION, THE CAUSE OF ONE FORM OF APPLE MEASLES[1]

JOHN C. DUNEGAN AND DWIGHT ISELY[2]

(Accepted for publication June 5, 1945)

INTRODUCTION

In 1912 Hewitt and Truax (5) reported the occurrence in northwest Arkansas of an apple bark disease which they named "measles." This disease had two distinct forms, the pustular type and the scurfy type. As no pathogenic fungus or bacterium was isolated from the affected tissues they considered the disease a physiological trouble.

In the pustular type, which they found to be the more prevalent form, the disease first appeared as " . . . scattered red pycnidia-like pustules on the young smooth bark. . . ." As the disease progressed the pustules became very numerous and the bark appeared very rough.

A histological study of these pustules revealed that " . . . they seemed to burrow into the sound tissue underlying them . . ." to varying depths depending upon the age of the pustule. The entire pustule appeared to be " . . . a mass of cork cells with the whole cell nearly filled with some substance which is yellowish brown . . ." while " . . . in the older pustules the tissue in the middle is disorganized leaving a hollow core. . . ."

In contrast to these pronounced histological changes asosciated with the pustular type, Hewitt and Truax found that in the scurfy type " . . . the phelloderm is irregularly thickened for a considerable area on the branch . . ." but " . . . the growth in the scurfy area is at no point as deep as in the pustules, the thickening of the phelloderm amounting to no more than six or eight cells in depth. . . ." Occasionally the two types were intermixed and then the affected area in the scurfy type was " . . . 5–20 cells deep with the deeper portions in specimens which contain pustules along with the scurf. . . ."

These excerpts from the original description show that the symptoms of the pustular type differ markedly from those of the scurfy type. Furthermore, the pustular type of measles, as described by Hewitt and Truax, is not identical with other obscure apple bark diseases such as the rough-bark or scurfy canker described by Rose (9), the isolated pustular, the aggregate pustular, and canker types described by Rhoads (7), target canker described by Roberts (8), or black pox and internal bark necrosis described by Berg (2). Likewise, the pustular type symptoms are not identical with the symptoms on apple bark resulting from boron deficiency observed by Young and

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Winter (11), Burrell (3), and Hildebrand (6), nor with those resulting from the effect of high soluble-salt concentration in the soil, described by Crawford (4) in New Mexico. This becomes especially apparent when it is realized that although Hewitt and Truax emphasized, both in their text and illustrations, the "burrowing tendency" and "hollow core" of the pustules, these striking histological characteristics are not stressed by any of these subsequent investigators. Nevertheless, specimens having all the morphological characteristics ascribed by Hewitt and Truax to the pustular type of apple measles can be found in the apple orchards of northwest Arkansas at the present time. A comparative study of this material is presented.

THE PUSTULAR TYPE OF APPLE MEASLES

The most logical procedure for such a comparative study would be one based on a comparison of present-day specimens with the original material collected by Hewitt and Truax. Unfortunately, none of the original specimens were found in the herbarium of the Department of Plant Pathology, University of Arkansas.[3] In lieu of the original specimens the writers based their study on the original description of the pustular type, as well as on unpublished photographs located during the search for the original specimens.

The photographs (Fig. 1, A to E from the files of the Department of Plant Pathology or reproduced from the original paper) illustrate the macroscopic and microscopic appearance of the pustular form of apple measles. Hewitt and Truax were able to strip off the burrowing pustules leaving holes (Fig. 1, D) in the cortex and the intact pustules (Fig. 1, E) attached to the phelloderm and epidermis.

The pustules found at the present time in apple twigs in northwest Arkansas orchards are hemispherical, from 0.25 mm. to 0.75 mm. in diameter, and are raised from 0.1 mm. to 0.25 mm. above the surface of the twig. Superficially they resemble fungus pycnidia, as Hewitt and Truax mentioned in their original account of the disease. In addition to these hemispherical pustules, elongate blister-like swellings are also present on the twigs. These elongate swellings were not mentioned by Hewitt and Truax, yet one of their photographs (reproduced as Fig. 1, A) shows the presence of elongate swellings on the twigs they studied.

In the present study both the pustules and the blister-like swellings began to appear on the current season's growth of a few twigs about the middle of August. In September and October pustules were present on practically all the new growth. They were more numerous on some twigs than on others, but their numbers did not increase after the first frost.

These pustules persist on the twigs for several years. The older pustules are larger, have a less regular outline, and have a slight depression at their center. These changes can be attributed to the subsequent growth of the twig, and it is this growth that makes the surface of the twigs very uneven

[3] Hewitt also advised the writers in 1938 that he had none of the original material.

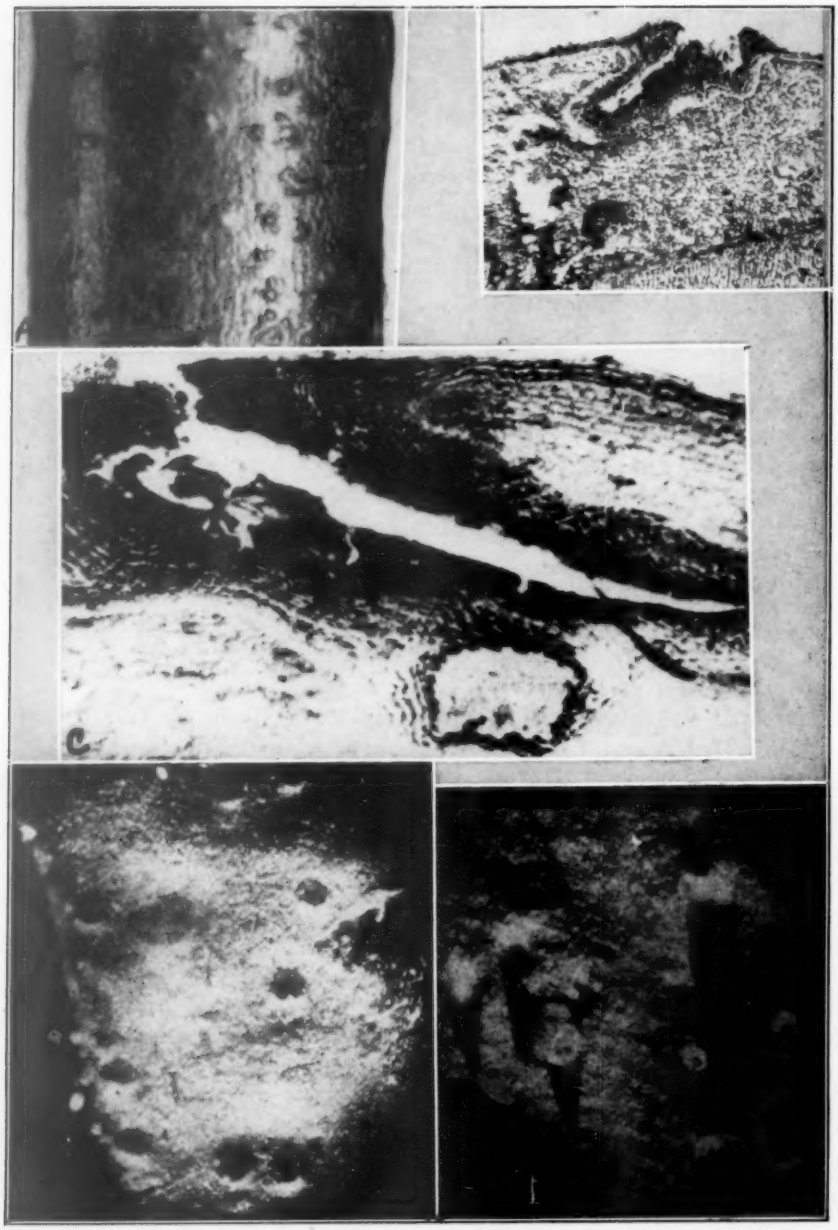

FIG. 1. A. Pustular type of apple measles. Print made from Hewitt's original negative in the files of the Dept. of Plant Pathology, University of Arkansas. B. "Section of an old pustule showing deep burrowing." Legend and figure reproduced from Fig. 8 of Bulletin 112, Arkansas Agricultural Experiment Station. C. "An older pustule showing diseased tissue surrounding a strand of bast." Legend and figure reproduced from Fig. 9 of Bulletin 112, Arkansas Agricultural Experiment Station. D. "Surface view of cortex showing the holes left by removing pustules." Legend and figure reproduced from Fig. 13 of Bulletin 112, Arkansas Agricultural Experiment Station. E. "Surface view of the underside of the phelloderm. The projections are the pustules extending inwards." Legend and figure reproduced from Fig. 12 of Bulletin 112, Arkansas Agricultural Experiment Station. All illustrations and legends in Fig. 1 are reproduced by permission of the Director of the Arkansas Agricultural Experiment Station.

as they grow older. Eventually the pustules lose their distinct outlines and are finally sloughed off, leaving areas of rough bark on these older branches.

CAUSE OF THE PUSTULAR TYPE

The first clue concerning the agent responsible for the pustular type of measles was obtained in 1936, when in examining small pustules on apple twigs received from Oregon it was found that a viscid liquid oozed from a number of the pustules as they were dissected under a binocular microscope. Further dissection revealed the presence in the center of each pustule of an insect egg, which was identified as that of a leafhopper. The twigs were placed in covered battery jars and in due course nymphs of a leafhopper, identified as *Typhlocyba pomaria* McAtee, the white apple leafhopper, emerged from the spots on the twigs.[4]

The elongate blister-like swellings which occur intermixed with the typical isolated pustules of apple measles in Arkansas are readily recognized as having been caused by the deposition of the winter eggs of the white apple leafhopper. Furthermore, the period of formation of the measles pustules coincides with the period of deposition of these eggs and suddenly ceases following the first frost. This suggested that leafhoppers might be involved also in the production of the pustules. The difference in appearance between the elongated blister-like swellings and pustules might be due solely to the angle at which the ovipositor was inserted in the twig.

Washburn (10) reported in 1908 that Ainsle and Webster, who were working with him in Minnesota, had found the winter eggs of a leafhopper in apple twigs during the early part of the 1907 growing season. He described and figured not only what is now regarded as the typical elongate swelling caused by leafhopper oviposition but also an almost "cylindrical pouch" similar to the measles pustules observed by the writers in Arkansas.

When pustules from Arkansas orchards were examined under a binocular microscope, a leafhopper egg was found embedded in each pustule. Histological changes in sections of these pustules were similar to those described and photographed by Hewitt and Truax, as a comparison between Fig. 1, B and C, and Fig. 2, A and B, shows.

These pustules, caused by the insertion of a leafhopper egg, had the "burrowing tendency" observed by Hewitt and Truax. The egg was in the central portion or core of the pustule in many of the sections.

Sections of these pustules on one-year-old and two-year-old wood showed even more clearly the "burrowing tendency" and the "hollow core," as well as the tendency for the spot to involve the pericyclic fibers (Fig. 2, C).

In 3-year-old and 4-year-old wood the subsequent growth of the twigs had produced further marked changes in the pustules themselves. The hollow core disappeared (Fig. 2, D) as the pressure from the twig growth forced the walls of the cavity together, and finally, in the oldest pustules examined, only a solid mass of discolored cells marked the site of the original egg cavity.

[4] The junior writer (1) had been engaged in a study of the biology of leafhoppers in the Ozarks for several years previously and had reared a number of series of *Typhlocyba pomaria* in all its stages.

It was difficult to detect fragments of the egg shell in cross sections of this mass of discolored cells, but by removing layer after layer of the sur-

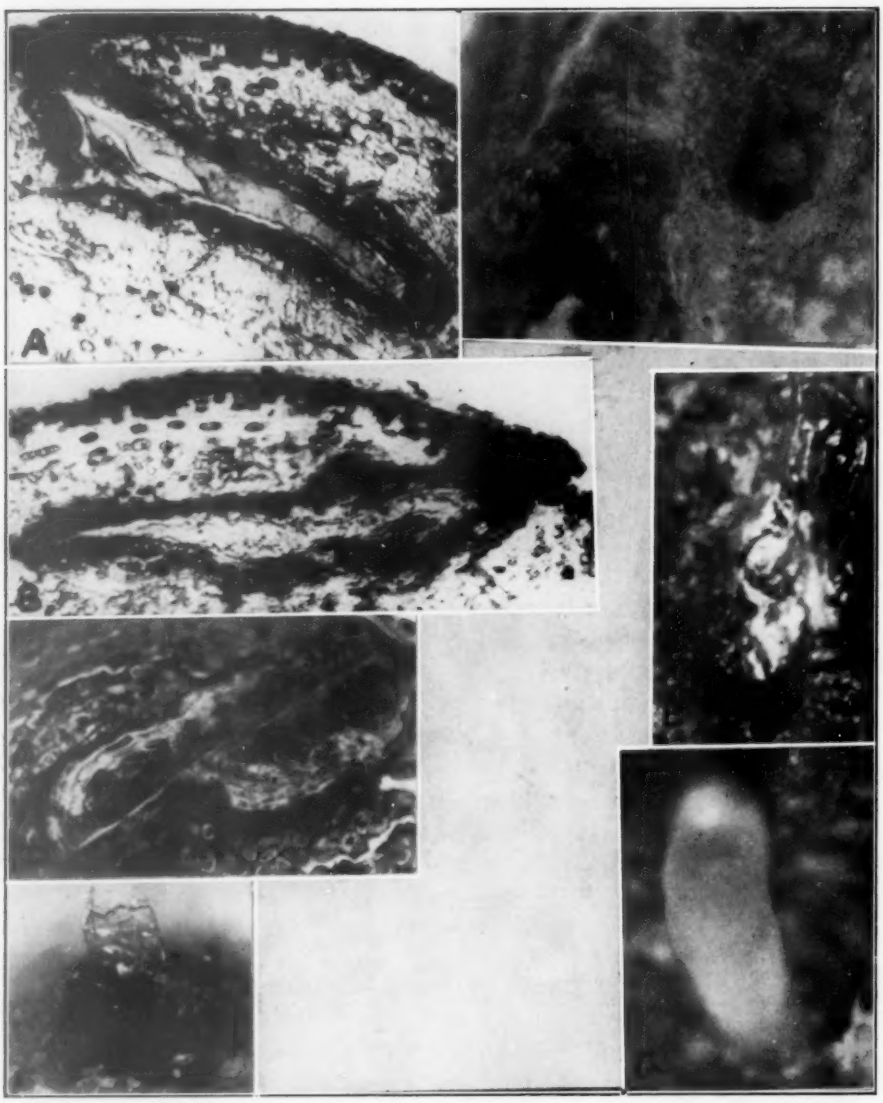

FIG. 2. A and B. Cross sections of pustules showing eggs of *Typhlocyba pomaria* embedded in tissues of Delicious apple twigs. Note the "burrowing tendency." C. Section showing pericyclic fibers adjacent to a pustule enveloped by a wound periderm. D. Section of older pustule showing the egg cavity being obliterated. E. Outer tissues of old pustule dissected away, revealing fragments of leafhopper egg in the central core of the pustule. F. Fragment of egg shell projecting from central core after surrounding tissues had been dissected away. G. Leafhopper egg attached to underside of epidermis after being withdrawn from young pustule.

rounding tissues under a binocular microscope it was possible to find portions of the egg shell (Fig. 2, E and F) in the center of the pustules.

In contrast to the difficulties encountered with the older pustules, it was relatively easy to withdraw the egg intact from the young pustules (Fig. 2, G), leaving visible a hole in the cortical tissues.

Hewitt and Truax were able to withdraw the entire pustule by soaking the twigs in an aqueous solution of sulphurous acid. Their illustration (reproduced as Fig. 1, E) shows dark spots in the center of a number of the pustules. The writers noted the same sort of dark spots in the center of the old pustules when the entire pustule was removed intact by dissection. Fragments of the leafhopper egg shell were found in these dark spots.

DISCUSSION

The evidence just presented suggests that the pustular type of measles originally described by Hewitt and Truax is caused by the deposition of eggs of *Typhlocyba pomaria*[5] in the tissues of apple twigs. Since the original specimens apparently are no longer in existence it is impossible to demonstrate that leafhopper eggs actually were deposited in the twigs from which Hewitt and Truax prepared their original description. However, this cause is indicated by the following circumstantial evidence: (1) Pustules similar to those described by Hewitt and Truax as the pustular stage of apple measles may be found on apple twigs in Arkansas orchards at the present time. (2) These pustules have the "burrowing tendency" and in the older stages have the "hollow core" described and illustrated by Hewitt and Truax in their original paper. (3) Leafhopper eggs or egg-shell fragments can be found in these pustules. (4) In microscopical sections of the pustules known to be the result of leafhopper egg-laying activities the histological changes are identical with those described and illustrated by Hewitt and Truax. (5) The pustular stage described by Hewitt and Truax is superficial, *i.e.*, the histological changes involve the epidermis, phelloderm, and outer layers of the cortex. Their illustrations show a definite opening in the epidermis connected with the hollow core or central portion of the pustule. This is precisely the condition to be expected if the pustule was caused by a leafhopper. The egg was inserted through the opening and the nymph likewise emerged through the opening. (6) Hewitt and Truax noted that the bast fibers (pericyclic fibers) were sometimes involved. The same phenomenon may be observed in sections of pustules containing leafhopper eggs. (7) Hewitt and Truax were able to withdraw the entire pustule attached to the epidermis after proper treatment to soften the tissues. The pustules studied by the writers can be removed intact by dissection, leaving holes in the cortex similar to those pictured by Hewitt and Truax. (8) Hewitt and Truax's illustration of the underside of the pustules shows dark spots in the center of the cores. The same sort of spots, containing remnants of the egg shell, were noted in the pustules removed intact by the writers.

[5] According to Ackerman and Isely (1) there are five species of leafhopper attacking the apple in northwest Arkansas, but only two of these species, namely, *Typhlocyba pomaria* and *Empoasca maligna*, deposit their eggs in the twigs. Of these *T. pomaria* is the only one numerous enough to be responsible for the symptoms discussed.

The similarity between the pustules described by Hewitt and Truax as a form of apple measles, and those caused by the egg-laying activities of *Typhlocyba pomaria* is sufficient, the writers consider, to prove that the two types of pustules are identical.

This being the case, it may be concluded that the pustular type of apple measles disease as originally described by Hewitt and Truax is not a "physiological trouble" but is caused by the egg-laying activities of the leafhopper *Typhlocyba pomaria*.

SUMMARY

Apple measles as originally described by Hewitt and Truax consisted of two types: the pustular and the scurfy.

The symptoms of the pustular type are distinctly different from those of the scurfy type and likewise are not identical with those of other obscure apple bark diseases described by later investigators.

A comparison of material collected in Arkansas in recent years with the original description and photographs (none of the original specimens being available) of the pustular type convinced the writers that this type still occurs in Arkansas orchards.

A study of these present-day specimens shows that the pustules are caused by the insertion of the winter eggs of the white apple leafhopper, *Typhlocyba pomaria* McAtee, in the twigs.

The histological features described and illustrated by Hewitt and Truax are found in apple twig tissues injured by leafhopper oviposition.

The writers conclude that the pustular type of measles, as originally described by Hewitt and Truax, is caused by the deposition of leafhopper eggs in the twigs.

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LITERATURE CITED

1. ACKERMAN, A. J., and D. ISELY. The leaf hoppers attacking apple in the Ozarks. U. S. Dept. Agr. Tech. Bull. 263. 1931.
2. BERG, A. Black pox and other apple-bark diseases commonly known as measles. W. Va. Agr. Exp. Stat. Bull. 260. 1934.
3. BURRELL, A. B. The boron-deficiency disease of apple. N. Y. (Cornell) Agr. Exp. Stat. Ext. Bull. 428. 1940.
4. CRAWFORD, R. F. Apple measles. N. Mex. Agr. Exp. Stat. Bull. 251. 1937.
5. HEWITT, J. L., and H. E. TRUAX. An unknown apple tree disease. Ark. Agr. Exp. Stat. Bull. 112. 1912.
6. HILDEBRAND, E. M. Internal bark necrosis of Delicious apple, a physiogenic "boron-deficiency" disease. (Abstr.) Phytopath. 29: 10. 1939.
7. RHOADS, A. S. Apple measles, with special reference to the comparative suceptibility and resistance of apple varieties to this disease in Missouri. Phytopath. 14: 289–314. 1924.
8. ROBERTS, J. W. Apple target canker, measles and rough bark. (Abstr.) Phytopath. 24: 16. 1934.
9. ROSE, D. H. Blister spot of apples and its relation to a disease of apple bark. Phytopath. 7: 198–208. 1917.
10. WASHBURN, F. L. Egg laying of *Empoasca mali*. Jour. Econ. Ent. 1: 142–145. 1908.
11. YOUNG, H. C., and H. F. WINTER. The effect of boron, manganese, and zinc on the control of apple measles. Ohio Agr. Exp. Stat. Bi-Mon. Bull. 188: 147–152. 1937.

GENETIC LEAF ROLL OF IRISH POTATO SEEDLINGS[1]

E. L. LeCLERG[2]

(Accepted for publication June 15, 1945)

A leaf roll of small Irish potato seedlings grown from true seed in 3-inch pots was observed in the greenhouse during the winter of 1943. It was first observed in the progeny of a self-pollinated seedling (XL 72–1) which had been selected in Louisiana. The margins of the leaves of the affected plants rolled upward starting with the lower leaves and extending to the apical leaves (Fig. 1). Many of the rolled leaves had a red-purple tinge along the margins shortly after rolling became apparent.

FIG. 1. Genetic leaf roll of potato seedlings. Normal plant in center.

Table 1 gives the number of segregates with leaf roll in progenies from crosses as well as in inbred progenies. It appears that 45 of 124 plants were rolled in the inbred progeny IL 12 (from XL 72–1 selfed). Leaf-rolled plants appeared in 4 out of 6 cross-progenies in which seedling XL 72–1 was used as a parent; however, in the other 2 cross-progenies (XL 225 and XL 237) no rolled plants occurred. The progeny XL 237 [(XL 72–1) × (XL 105–2)] consisted of 1,477 segregates, which is a number sufficiently large to indicate that some factor may be operating to inhibit expression of this character in the segregates of this progeny.

Stem-graft inoculations with these rolled plants on disease-free stocks all gave negative results, which indicates that the rolling was not caused by a virus. Tubers produced by plants subject to rolling were planted at Crossville, Tennessee, in the spring of 1944. None of the plants produced from these tubers showed any type of leaf rolling during the growing season.

[1] These investigations were conducted by the Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering; Agricultural Research Administration, United States Department of Agriculture, in cooperation with the Department of Horticultural Research of the Louisiana Agricultural Experiment Station.

[2] Pathologist, Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering; Agricultural Research Administration, United States Department of Agriculture; stationed at Baton Rouge, Louisiana.

Had the rolling of the leaves in the seedlings been caused by a virus the plants grown in the field from these tubers also should have been rolled; accordingly, the fact that no rolling occurred in the field-grown plants is further evidence that a virus is not associated with this type of leaf rolling. Furthermore, it is apparent from these results that this type of leaf roll is different from the nonvirus leaf roll previously described by the writer.[3] The leaf roll herein discussed is limited to the small seedlings grown from true seed in clay pots and is not present in plants grown from tubers produced by these plants. The nonvirus leaf roll previously described, on the other hand, occurs only in field-grown plants and not in small seedlings.

TABLE 1.—*Expression of genetic leaf roll in potato seedlings grown from true seed in the greenhouse at Baton Rouge, Louisiana, in 1943*

Pedigree number	Parentage	Number of	
		Rolled plants	Nonrolled plants
XL 225	Katahdin × (XL 72–1)	0	34
XL 226	Sebec × Katahdin	0	28
XL 227	Katahdin × Sebec	0	25
XL 228	Sebec × (XL 72–1)	9	330
XL 229	(XL 72–1) × Sebec	8	287
XL 230	528–170 × (XL 72–1)	16	620
XL 232	(XL 72–1) × 528–170	5	172
XL 233	(XL 105–2) × Katahdin	0	925
XL 235	Friso × Earlaine	0	17
XL 236	Friso × Katahdin	0	24
XL 237	(XL 72–1) × (XL 105–2)	0	1,477
XL 238	Katahdin × 528–170	0	80
XL 239	528–170 × Katahdin	0	322
XL 240	(XL 105–2) × 528–170	0	1,273
IL 10	Sebec selfed	0	60
IL 11	528–170 selfed	0	752
IL 12	XL 72–1 selfed	45	124
IL 13	Katahdin selfed	0	133
IL 14	Earlaine selfed	0	132

The results herein reported support the hypothesis that this type of leaf rolling is the result of the expression of a genetic factor.

Katahdin, Earlaine, and Sebec do not possess factors for this type of rolling since the character did not appear in inbred progenies of these varieties (Table 1). Likewise, seedling 528–170 did not have rolled plants in its inbred progeny.

It has been impossible to formulate a genetical factorial explanation of this character from the data in table 1. However, this type of leaf rolling is due to a recessive factor.

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BATON ROUGE, LOUISIANA

[3] LeClerg, E. L. Nonvirus leaf roll of Irish potatoes. Amer. Potato Jour. 21: 5–13. 1944.

PRELIMINARY STUDIES ON PHYSIOLOGIC SPECIALIZATION IN TILLETIA TRITICI AND T. LEVIS IN CHINA[1]

T. F. Yu, H. R. Wang, C. T. Fang, and S. Y. Yin[2]

(Accepted for publication June 14, 1945)

INTRODUCTION

Stinking smut is one of the most important diseases of wheat in North China. It is so serious that, in certain spring wheat growing regions, wheat has been replaced by barley. In South China, the disease seems less prevalent and destructive. Nevertheless epidemics of bunt have frequently been reported from certain localities.

The best method of reducing loss from stinking smut is the development of smut-resistant varieties. However, the problem of producing smut-resistant varieties is complicated by the existence of many physiologic races within *Tilletia tritici* (Bjerk.) Wint. and *T. levis* Kuhn. For this reason, varieties resistant to certain races may remain resistant only as long as they are exposed to infection by only such particular physiologic race or races of the pathogens. It seems, therefore, that in the course of developing stinking smut resistance in wheat, it is highly desirable to begin with studies on the number, prevalence, distribution, and pathogenic capability of physiologic races.

LITERATURE REVIEW

The first statement that specialization occurs in bunt was made by Faris (4), who, in the course of investigating the physiologic factors influencing infection by stinking smut organisms, obtained some preliminary evidence of differences in pathogenicity. Since then, specialization in both *Tilletia tritici* and *T. levis* has been studied by many investigators (1, 2, 3, 5 through 15), who have shown conclusively that these smut fungi comprise distinct physiologic races.

Because of the lack of uniformity in the use of differential hosts and in the system of numbering distinct races, many of the races identified and numbered by the various workers might be duplicates. In an attempt to work out a standard system for the identification of physiologic races of bunt organisms in the United States and Canada, Rodenhiser and Holton (16) have reinvestigated the races described by the previous workers and, in addition, have tested a number of collections of smut. They have selected 10 varieties of both winter and spring wheats as the standard differential hosts and were able to identify and number 10 races of *T. tritici* and *T. levis*.

[1] Paper No. 22 from Division of Plant Pathology, The Institute of Agricultural Research, National Tsing Hua University.

[2] The writers wish to express their appreciation to Prof. F. L. Tai for his encouragement throughout the investigation and his criticism of the manuscript and to Dr. E. C. Stakman for reading the manuscript. They are also indebted, for bunt material, to Dr. C. S. Wang, Honan; Mr. T. L. Nien, Kweichow; and Dr. L. Ling and Mr. Y. S. Wu, Szechwan. Dr. H. A. Rodenhiser of United States Department of Agriculture very kindly supplied seed of the differential wheat varieties.

Recently, Holton and Rodenhiser (12) have described 5 new races of *T. tritici* and 2 of *T. levis*. In this study, Martin and White Odessa were added as differential hosts in the winter wheat group, while Mindum was omitted, as a differential host, from the spring wheat group.

MATERIALS AND METHODS

Collections of bunt were requested in 1938 and 1939 from many of the agricultural institutes in China. Forty-five collections were obtained for use as inoculum in the fall of 1938 and 12 more were obtained in the fall of 1939. Most of these collections used in the present studies were from single galls and were propagated on susceptible wheat. Every precaution was taken to keep them as pure as possible.

The smut reactions of 24 varieties of wheat were studied. Of these wheats 14 were either developed or introduced from foreign countries by the various agricultural experiment stations in China and the remaining 10 were those used by Rodenhiser and Holton for differentiating the physiologic races of the two species of *Tilletia* in the United States. Most of the Chinese wheats, with the exception of 2H80, which was one of Percival's wheats introduced into China, failed to differentiate the smut collections thus far obtained. Among the wheats selected by Rodenhiser and Holton as their differential hosts, Ulka and Hybrid 128 were susceptible and Ridit, Oro, Hussar, and Canus were resistant to all of the smut collections in China. Albit did not grow well, and Hohenheimer was discarded on account of its high degree of sterility under local conditions. Thus, only three varieties of wheat, Marquis, Mindum, and 2H80, finally were selected as differential hosts.

Seeds of differential hosts were treated with formaldehyde according to the standard method, thoroughly washed with water, and allowed to dry. The seeds and spores were then shaken together in an envelope until the seed was completely covered with spores. Three grams of inoculated seeds were planted in each 3-ft. row. Three rows constituted a plot, and plots were systematically arranged in 2 or 3 replications. Smut percentages obtained were based on counts of the total number of heads per plot. Three infection classes were used: Resistant class (R)—0–5 per cent infection; intermediate class (I)—6–20 per cent infection; and susceptible class (S)—21–100 per cent infection.

RESULTS

The results of 3 years' work are given in table 1. Reactions of the varieties were remarkably consistent. Ulka, which was used by Rodenhiser and Holton as the susceptible check, was also extremely susceptible to all of the smut races in China. Its smut reactions are in table 1 for comparison with reactions of the 3 differential hosts.

Nine of the 57 smut collections were *Tilletia levis*. On the basis of the percentages of infection they produced, they represent 6 distinct races.

Race 1 is the only race to which the 3 differential hosts are resistant. Race 2 is differentiated from race 1 by its capacity to infect Mindum, and race 6 by its development on Mindum and 2H80. Marquis has an intermediate reaction to race 3, and 2H80 has an intermediate reaction to race 4. Race 5 differs from race 4 by the intermediate reaction of Mindum and from race 3 by the intermediate reactions of both Mindum and 2H80.

A total of 48 collections of *Tilletia tritici* have been tested. On the basis of reactions of the same three differential hosts, these collections comprised 4 distinct races. All three differential hosts are resistant to race 1. Race 2 is differentiated from race 1 by the susceptible reaction of Mindum, race 3

TABLE 1.—*Percentages of bunt obtained in 3 varieties of wheat exposed to 4 physiologic races of Tilletia tritici and 6 physiologic races of Tilletia levis at Kunming, China*

Host variety	Year tested	Percentages[a] of smutted heads produced by									
		Races of *T. tritici*				Races of *T. levis*					
		1	2	3	4	1	2	3	4	5	6
Marquis	1941	2.8	1.1	10.1	3.7	3.7	3.7	11.7	8.2	5.8	7.2
	1942	0.8	0.6	6.4	12.9	1.2	2.7	20.2	9.0	5.3	4.7
	1943	0.5	0.6	7.7	5.3	1.5	3.5	5.0	13.7	7.6	6.1
	Av.	1.4	0.8	8.1	7.3	2.1	3.5	12.3	10.3	6.7	6.1
Mindum	1941	0.9	18.5	5.3	2.0	0.0	20.0	0.0	3.6	5.2	11.2
	1942	1.1	24.2	3.1	6.6	0.0	20.8	0.0	0.9	7.2	8.4
	1943	0.0	22.6	3.7	1.6	0.0	25.3	0.0	4.1	5.7	9.0
	Av.	0.7	21.8	4.0	3.4	0.0	22.2	0.0	2.9	6.1	6.2
2H8O	1941	0.0	0.4	0.0	5.1	0.0	0.0	0.0	6.1	6.3	17.9
	1942	0.0	0.0	0.0	13.0	0.0	0.0	0.0	9.4	3.3	15.4
	1943	0.0	0.0	0.0	2.7	0.0	0.4	0.0	7.6	5.7	34.8
	Av.	0.0	0.1	0.0	6.9	0.0	0.1	0.0	7.7	5.1	24.0
Ulka	1941	23.4	45.2	43.7	47.1	41.3	24.0	47.5	73.6	48.8	63.1
	1942	45.5	47.8	52.1	35.5	29.8	43.5	51.2	59.8	43.0	49.9
	1943	35.7	49.8	39.0	64.3	31.0	26.5	30.6	45.7	47.7	43.7
	Av.	34.9	45.9	44.9	49.3	34.0	31.3	43.0	59.7	46.6	55.6

[a] Percentages for each year are averages from 3 or 4 plots.

by the intermediate reaction of Marquis, and race 4 by the intermediate reaction of 2H80. Table 2 and the keys show that the 6 races of *T. levis* and 4 races of *T. tritici* may be differentiated with the three varieties of wheat used.

Of the 9 smut collections obtained from North China, 7 were *Tilletia levis*. This probably indicates that *T. levis* is the predominant species in North China. Of the 48 smut collections made in South China, however, only 2 were identified as *T. levis*. Evidently, in South China *T. tritici* is predominant. The number of collections of *T. levis* was too small to permit a detailed analysis of the distribution of the species in China but the 45 collections of *T. tritici*, comprising 4 distinct races, may give a general idea of the distribution and prevalence of these races and of the species in South-

western China. The frequency of occurrence and the distribution by provinces and districts of the collections representing the 4 races of *T. tritici* are in table 3.

TABLE 2.—*Reactions of 3 varieties of wheat which differentiate 6 physiologic races of Tilletia levis and 4 races of Tilletia tritici*

Host variety	Race No. of *T. tritici*				Race No. of *T. levis*					
	1	2	3	4	1	2	3	4	5	6
Marquis	R	R	I	I	R	R	I	I	I	I
Mindum	R	S	R	R	R	S	R	R	I	I
2H8O	R	R	R	I	R	R	R	I	I	S

Race 1 of *Tilletia tritici* was collected more often than all other races and it occurred over wide areas in Southwestern China. Race 4 was collected only once in Kwangshung, Kweichow.

Key to 4 physiologic races of Tilletia tritici

	Race No.
Marquis resistant	
Mindum resistant	1
Mindum susceptible	2
Marquis intermediate	
2H8O resistant	3
2H8O intermediate	4

Key to 6 physiologic races of Tilletia levis

	Race No.
Marquis resistant	
Mindum resistant	1
Mindum susceptible	2
Marquis intermediate	
Mindum resistant	
2H8O resistant	3
2H8O intermediate	4
Mindum intermediate	
2H8O intermediate	5
2H8O susceptible	6

SUMMARY AND CONCLUSIONS

Three years' data on physiologic specialization in *Tilletia tritici* and *T. levis* in China have been obtained. Separation of races within each species was based on differences in pathogenicity on three varieties of wheat, Marquis, Mindum, and 2H8O. Four races of *T. tritici* and 6 races of *T. levis* were identified and numbered T–1 to T–4 and L–1 to L–6 respectively.

In North China *T. levis* strongly predominates while in South China *T. tritici* is predominant. Race 1 of *T. tritici* is the most widely distributed in Southwestern China. It was found in 17 of the 18 districts from which collections were obtained. It was also the most frequently collected, comprising 35 of a total of 45 collections. This race, to which all the differential hosts are resistant, may be further separable into more races if the proper additional differential hosts are used.

In the northwestern corner of Yunnan, where bunt is usually epidemic, races 1, 2, and 3 of *Tilletia tritici* have been thus far found. The wheat

TABLE 3.—*Frequency of occurrence and distribution by provinces and districts of collections of 4 physiologic races of Tilletia tritici in Southwestern China*

Province and district	No. of collections of each race				Total collections
	1	2	3	4	
Yunnan					
Tali	2	1	.....	.....	3
Hsihchow	1	.....	2	.....	3
Tungchwan	2	1	.....	.....	3
Feng-I	3	.....	.....	.....	3
Yang-pi	4	1	.....	.....	5
Tengschwan	2	.....	.....	.....	2
Total	14	3	2	.....	19
Kweichow					
Ping-I	2	.....	.....	.....	2
Ping-pa	3	.....	.....	.....	3
An-shun	3	.....	.....	.....	3
Tsingcheng	2	.....	.....	.....	2
Kwangshung	1	.....	.....	1	2
Chengning	.....	1	.....	.....	1
Total	11	1	.....	1	13
Szechwan					
Sui-ning	2	2	.....	.....	4
Si-chung	3	1	.....	.....	4
Feihsien	2	.....	.....	.....	2
Chingtang	1	.....	.....	.....	1
Anhsien	1	.....	.....	.....	1
Santai	1	.....	.....	.....	1
Total	10	3	.....	.....	13
Grand total	35	7	2	1	45

variety 2H80 has been resistant to these 3 races. This wheat possesses, in addition to its smut resistance, many other desirable agronomic characteristics and will be valuable in breeding wheat resistant to stinking smut for this part of China.

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LITERATURE CITED

1. AAMODT, O. S. Varietal trials, physiologic specialization, and breeding spring wheats for resistance to Tilletia tritici and T. levis. Can. Jour. Res. **5**: 501–528. 1931.
2. BRENTZEL, W. E. Physiologic specialization of Tilletia tritici on emmer. Phytopath. **23**: 483–485. 1933.
3. BRESSMAN, E. N. Varietal resistance, physiologic specialization and inheritance studies in bunt of wheat. Oreg. Agr. Exp. Stat. Bull. 218. 1931.
4. FARIS, J. A. Factors influencing the infection of wheat by Tilletia tritici and Tilletia levis. Mycol. **16**: 259–282. 1924.
5. FLOR, H. H. Heterothallism and hybridization in Tilletia tritici and T. levis. Jour. Agr. Res. [U.S.] **44**: 49–58. 1932.
6. ———. Studies on physiologic specialization in Tilletia tritici and Tilletia levis in the Pacific Northwest. Jour. Agr. Res. [U.S.] **47**: 193–213. 1933.
7. GAINES, E. F. New physiologic forms of Tilletia levis and T. tritici. Phytopath. **18**: 579–588. 1928.
8. GASSNER, G. Über Auftreten und Verbreitung von Tilletia tritici und Tilletia foetans in der Türkei. Phytopath. Ztschr. **11**: 468–488. 1938.

9. HOLTON, C. S. The probable explanation of recent epidemics of bunt in durum wheats. Phytopath. **20**: 353–357. 1930.
10. ————. The relation of physiologic specialization in Tilletia to recent epiphytotics of bunt in durum and Marquis wheats. Phytopath. **21**: 687–694. 1931.
11. ————. Studies of seven differentiating characteristics of two physiologic forms of Tilletia tritici. Phytopath. **25**: 1091–1098. 1935.
12. ————, and H. A. RODENHISER. New physiologic races of Tilletia tritici and T. levis. Phytopath. **32**: 117–129. 1942.
13. MELCHERS, L. E. Investigations on physiologic specialization of Tilletia levis in Kansas. Phytopath. **24**: 1203–1226. 1934.
14. REED, G. M. Physiologic races of bunt of wheat. Amer. Jour. Bot. **15**: 157–170. 1928.
15. RODENHISER, H. A., and E. C. STAKMAN. Physiologic specialization in Tilletia tritici. Phytopath. **17**: 247–253. 1927.
16. ————, and C. S. HOLTON. Physiologic races of Tilletia tritici and T. levis. Jour. Agr. Res. [U.S.] **55**: 483–496. 1937.

EPIDEMIOLOGY STUDIES ON STRIPE RUST OF WHEAT IN CHENGTU PLAIN, CHINA

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Stem, leaf, and stripe rusts are all of common occurrence on wheat in Szechuan province, China, but stripe rust is often the most important factor in the reduction of wheat yields. Although this rust is seldom absent, it varies considerably in severity from year to year. In Chengtu plain of this province, the rusts occur in the following sequence: (1) *Puccinia triticina* Erikss., (2) *P. glumarum* (Schmidt.) Erikss. and Henn., (3) *P. triticina* Erikss., (4) *P. graminis* Pers. In general, wheat is sown in that region in late October and harvested in the middle of May. Leaf rust appears on seedlings of early sown wheat about the middle of November, when the temperature averages not lower than 10° C. It is seldom seen when temperatures are lower. Stripe rust occurs in late November or early December and maintains itself without much spread until February of the following year, when it becomes epidemic if environmental conditions permit. In average years, the stripe rust disappears in late April. Leaf rust reappears in late March to early April and becomes prevalent in May. Stem rust is the last to appear and gains in intensity in June, but it is too late to cause damage on wheat.

NATURAL HOSTS

Besides attacking wheat and barley, stripe rust occurs very commonly on *Agropyron ciliare* (Trin.) Franch. and *A. semicostatum* Nees (probably identical with *A. caninum* (L.) Beauv.) in Chengtu plain and its surrounding mountains. On these two grass hosts, however, the rust appears much later than on wheat, generally not until April, and it becomes prevalent in May. It seems to endure much higher temperatures than stripe rust on wheat. Seedlings of *Bromus japonicus* Thumb. have been observed infected with stripe rust, but never mature plants. Other wild grasses such as *Bromus remotiflorus* Steud., *B. tectorum* L., and *Elymus sibiricus* Roch. have never been found infected. Rye is not cultivated in the plain region, but in the experimental plots it has not had stripe rust and has been seen rusted only by *Puccinia dispersa* Erikss. and Henn.

Since Newton and Johnson (12) and Becker and Hart (2) have found that stripe rust from *Agropyron* is capable of attacking wheat, the local Chinese collections from *Agropyron ciliare* were inoculated on the standard assortment of differential hosts to determine whether the physiologic races harbored on the grass host were identical with those on wheat. Two races were isolated from such collections. One attacked only two barley varieties, Fong Tien and Heils Franken, and produced scattered infections of type 2 instead of the usual stripe-like sori. The other attacked the two barleys

with type 3–4 infection, Petkuser rye with type 2 infection, and Chinese 166 wheat with type 3 infection. The latter race appears to be similar to 376A of Becker and Hart (2) from barley. It is also important to note that in 1941 the inoculations were made near the end of April and sori appeared in early May, a period during which the mean temperature was 24.7° C. At the same time several collections of rust from wheat were inoculated but failed to produce infections, apparently because of the high temperatures prevailing. Such results indicate that the grass is not likely to serve as a source of supply of urediospores for wheat during the unfavorable months in summer under local conditions.

EFFECT OF TEMPERATURE ON GERMINATION OF UREDIOSPORES

The cardinal temperatures for germination of urediospores of *Puccinia glumarum* have been studied by several investigators (7, 12, 14, 15). The minimum temperature is just above 0° C., the optimum ranges from 10° to 12° C., and the maximum from 23° to 27° C.

TABLE 1.—*The effect of temperature on the germination of urediospores of Puccinia glumarum*

Temperature, in degrees C.	Number spores counted	Germinated spores[a]	
		Number	Per cent
4.5	635	72	11.3
10	1,181	300	25.4
11.5	1,048	287	27.4
14	891	223	25.0
16	1,055	222	21.0
18	1,377	86	6.3
21	1,530	22	1.5
25	1,600	6	0.4
28	2,000	0	0
32	2,000	0	0

[a] After 18 hours.

As stripe rust is known to be particularly sensitive to high temperatures, the knowledge regarding the influence of temperature on the germination of urediospores of local physiologic races would be helpful in explaining the seasonal occurrence of stripe rust as well as the development of epidemics in China. In experiments in 1940, we used a race prevalent on native wheat which is similar to race 31 of Straib (14) except that it produces a 4 infection type on Spaldings Prolific wheat. The urediospores were shaken from the open pustules of infected leaves onto plates of potato-dextrose agar. The plates were immediately placed in incubators or in the refrigerator and examined at intervals. The average results of two experiments are summarized in table 1. At the optimum temperature of 11.5° C. germination started in four hours, reached over 1 per cent after 6 hours, and reached 27 per cent after 18 hours. The maximum appeared to lie around 25° C. The figures in table 1 are the averages obtained in two experiments.

LONGEVITY OF UREDIOSPORES

The profound influence of environmental conditions on the viability of urediospores of stripe rust is well indicated by the divergent results given by different investigators. Hungerford (6) found that the urediospores may remain viable for at least 58 days at ordinary room temperature in Oregon, United States, and that only a small percentage germinated after 63 days in a desiccator. In England, Mehta (7) obtained 5 per cent germination in the laboratory and 15–20 per cent at 2.5° or 5° C. after the urediospores had been exposed for one month. Ducomet (4), in France, determined that the urediospores were able to germinate during a period of at least of 235 days after formation. Becker (1) reported the retention of germinability of urediospores for 433 days at optimum conditions of 0° C. and 40 per cent relative atmospheric humidity. Menacacci (11) recorded that the urediospores taken from June infection in the vicinity of Rome and placed indoors lost their germinability on the 22nd day. Raeder and Bever (13) proved that the optimum humidity and temperature for the retention of germinability of urediospores were 49 per cent and 9° to 13° C. respectively. Under such conditions, spores remained viable for 88 days. The upper thermal limit of endurance for the rust was studied by Butler and Hayman (3), who found that the urediospores when moist would not endure exposure at 45° C., for 5 minutes.

Since the stripe rust apparently depends entirely upon the uredial stage for its perpetuation, the longevity of urediospores under local conditions was studied. As the urediospores found between the glumes have been reported to retain their germinative power longer (11), both the infected leaves and heads were used as the source of urediospores in the experiments of 1940. The viability of urediospores was tested at intervals by germinating them on agar plates. The materials were kept in open vials and held in the following conditions: (1) Outdoors, (2) in the laboratory, (3) in a desiccator, (4) in an incubator at 32° C. While the experiments were made the room temperature was approximately 20° C. and relative humidity was high.

Rusted leaves of a very susceptible native variety of wheat were collected on April 23. The initial percentage of germination of urediospores was 10.6. On April 27, the germinative power had decreased only slightly, except for those spores kept at 32° C., only 1.7 per cent of which germinated. After April 27, the germinability declined rapidly. On May 14, 21 days after the beginning of the experiment, the germination of urediospores kept outdoors was 3.2 per cent, while germinability of those under other conditions had decreased to little more than 1 per cent. On May 27, very few spores germinated under all the conditions tested.

Urediospores between glumes of Blé rouge prolifique barbu were collected April 27, and 21 per cent germinated. On May 3, the percentages of germination were all above 12 except for spores kept at 32° C., only 1.4 per cent of which germinated. On May 14, seventeen days after collection,

the germination decreased to 2.4 per cent for the urediospores held outdoors, to about 1 per cent for those in the laboratory and in the desiccator, and to a trace for those at 32° C. On May 27, few spores, either outdoors or in the desiccator, germinated, while none germinated in other treatments.

The results were checked occasionally by inoculating the urediospores directly to wheat seedlings. In all cases, the infectivity could be retained no longer than one week. These results agree with the fact that during the work of determining physiologic races, the rust samples sent from nearby districts through the mails usually failed to produce infections.

In 1943, similar experiments were made with urediospores from infected leaves kept in ordinary room conditions and outdoors. Spore germination was rather erratic, but, in general, the longevity of urediospores was about 18 days only.

In 1940, the effect of a constant high temperature of 36° C., which might be encountered locally in the hot summer, on the viability of urediospores of stripe rust was studied. At such high temperature, spores remained viable for only two days.

The urediospores of stripe rust are short lived under natural conditions, indoors or outdoors, or when kept in a desiccator. In the years tested, urediospores from infected leaves retained viability no longer than one month, while the urediospores between glumes were even shorter lived. The urediospores rapidly perished at high temperatures.

OVERSUMMERING OF STRIPE RUST

In the Chengtu plain, the winter is ordinarily so mild that the temperature seldom drops below 0° C. Therefore the stripe rust urediospores and mycelium overwinter without difficulty. Between the wheat harvest in May and sowing in October, however, there are about five months, part of which is a hot and wet summer. In July and August, the rainfall is heavy and temperatures above 30° C. frequently persist for several days. The relative atmospheric humidity is usually above 80 per cent during the summer and fall. Under such conditions, urediospores are unable to live long, and neither volunteer wheat nor *Agropyron* has been observed infected. Accordingly, the means by which the rust bridges over the gap between wheat crops must be sought outside the plain.

The Chengtu plain is bordered at its western edge by mountains of varying height. The peaks and ranges northwest of the plain are from 5,000 to 7,000 meters high (Fig. 1). Between low and high altitudes there is well-marked "vertical zonation" of climate and vegetation. The limit of growth of wheat and barley is approximately 3,600 m. (16). In the subalpine zone (3,000–5,000 m.), winter wheat is cultivated and its growing season is prolonged with the increase of elevation. At higher levels in this zone, wheat is sown in September and harvested the next August. Thus from the plain up to the high mountains, wheat is seen all the year around. The gap between crops in the plain is well bridged over by the croppings at higher altitudes.

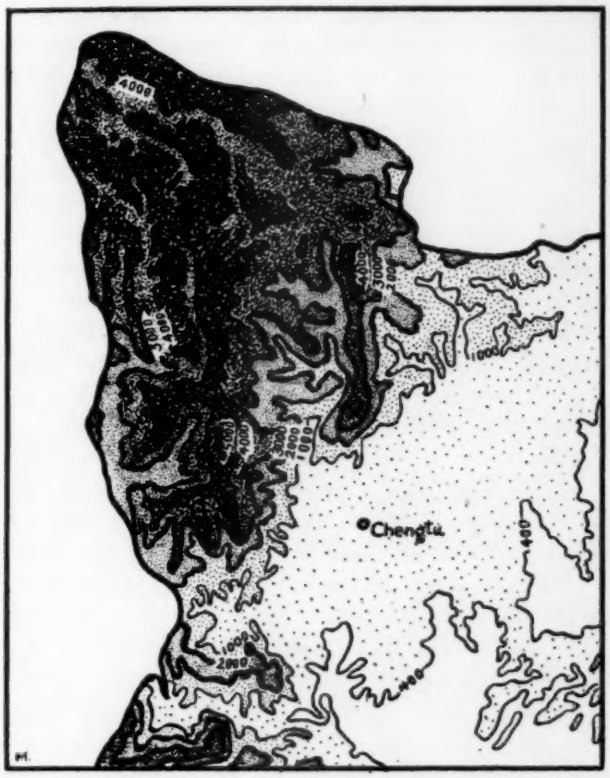

FIG. 1. Relief map of northwestern Szechuan Province, China.

In the plain, the stripe rust appears on wheat during May. Northwestward and in the mountain regions, the rust appears much later and may thrive throughout the summer. The monthly records of temperature in Sungpan (2,385 m.) as compared with those in Chengtu (503–576 m.) are in table 2. The rainfall on the mountains is always higher than in level country, which, in combination with lower summer temperatures, provides favorable conditions for rust development. Observations support such conclusions. At about 1,000 m. on Chineehnshan near Kwanhsien, stripe rust usually first appears in early April and is still seen in July on self sown wheat. In Lifan and Sungpan, at altitudes around 2,000 m., the stripe

TABLE 2.—*Average temperatures*[a] *at Chengtu and Sungpan, Szechuan Province, China*

Locality and altitude	Mean temperature, in degrees C.											
	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
Chengtu (503–576 m.)	5.9	8.4	12.5	17.3	22.1	24.9	26.8	26.2	22.0	18.1	12.2	8.1
Sungpan (2385 m.)	−1.7	−0.3	4.0	7.8	10.6	12.9	16.1	16.0	12.1	8.0	2.3	−1.7

[a] Averages for 1933–1943 at Chengtu and for 1937–1943 at Sungpan.

rust becomes prevalent in late June or July but is seldom seen in early August. In the spring wheat region on mountains near Sungpan, at altitudes slightly less than 3,000 m., the rust is not noticeable until the middle of July and it persists until harvest.

The evidence indicates that stripe rust survives the summer at higher altitudes. Above 2,500 m., there exists a gap of about one month between wheat crops. Since temperatures at altitudes above 2,000 m. seldom reach as high as April temperatures in the plain, there probably would be no difficulty for the urediospores to survive one month. Otherwise a quantity of urediospores from the spring wheat would supply sufficient inoculum for the fall crop at lower levels. According to the meteorological records of Sungpan, a northwest wind is not uncommon during October and November. The urediospores of the rust can be carried southeastward to the plain by the air current. In 1938, spore traps set in Chengtu first revealed the presence of urediospores of stripe rust in the air on November 28. Sporadic infections during the early season on wheat seedlings in the plain, which serve as the infection centers for further spread of the rust, also furnish evidence that inoculum is wind-borne. During April and May southeast winds and vertical movement of the air are frequent in the plain and doubtless facilitate the dissemination of urediospores to higher altitudes. The situation is comparable to that in India (8, 9, 10).

It is not known up to what levels the urediospores of stripe rust are able to survive the cold winter. According to Eriksson (5), they may endure $-4.5°$ to $-10°$ C. for 2 hours. But in the meteorological records of Sungpan (2,385 m.) temperature was as low as $-17.6°$ C. in January, 1941. However, on the high mountains there are likely to be early snows, little alternate thawing and freezing, and late spring thaws. From the records on the tolerance for low temperatures by other cereal rusts, there is no reason to expect that stripe rust is unable to overwinter in its uredial stage or by dormant mycelium at least as high as where wheat grows.

INFLUENCE OF CLIMATIC FACTORS ON THE DEVELOPMENT OF EPIDEMICS OF STRIPE RUST

The prevalence of stripe rust on wheat has been investigated at Chengtu from 1938 to 1944 and its correlation with the meteorological factors has been attempted. In the seven years, the worst epidemic occurred in 1939. There was comparatively little rust in 1943 and 1944. The other years can be classed as intermediate in rust severity. In general, the winter in Chengtu plain is mild and fairly dry, but not rainless; yet winter drought is not usually suffered, because the humid atmosphere and the cloudiness reduce evaporation. Severe frosts are rare. In spring the rainfall is much increased and the temperature is higher. Such conditions permit the safe overwintering and early outbreak of the rust in average years. Meteorological data during the critical period of rust development are summarized in table 3. The situation of each year is given briefly as follows.

TABLE 3.—*Meteorological records at Chengtu*

Year	Rainfall, in mm.		Relative humidity, in per cent (Feb.–Apr.)	Mean temperature, in °C. (Apr.)	Number of days of rain (Feb.–Apr.)
	Winter[a]	Spring			
1938	66.7	148.2	77.4	17.5	21
1939	29.6	175.6	81.3	14.1	40
1940	7.6	264.6	74.2	16.3	33
1941	29.0	118.5	71.5	19.5	24
1942	15.8	176.2	80.1	18.2	32
1943	13.0	91.1	77.2	16.8	24
1944	21.7	106.8	76.2	17.5	38

[a] Winter refers to December of the previous year and January and February of the year indicated.

In 1938, an intermediate rust year, the especially wet winter and the high rainfall in February brought about an early outbreak of rust in the middle of February. But the spring was relatively dry in comparison with an average year, therefore the rust was unable to reach its maximum destructiveness in April and became almost unnoticeable toward the end of April.

In 1939, an epidemic year, the active development of rust occurred a little later than in 1938. Although the rainfall was not particularly high in the spring, it was well distributed. Several other factors were in extremes in comparison with other years: 1939 had the highest relative humidity, the highest number of rainy days, the lowest number of hours with sunshine, and the lowest mean temperature in April. The cloudy and wet weather brought about a very succulent growth of wheat, on which the water was amply retained. Moreover, the maturity of wheat as well as the appearance of the telial stage of rust was delayed by the low temperature in April. The uredial stage therefore had excellent chances for reproduction and there was a wide and rapid spread of the rust. Throughout the period the rust had an unusually thriving development until the temperature suddenly rose in early May. In the wheat fields the soil was covered by a yellow layer of spores for more than two months. This year was also very favorable to the growth of wheat, but many varieties failed to yield well because of the severe rust attack.

The intermediate rust year, 1940, was a year with weather conditions roughly contrary to 1938. The winter was very dry but the spring was wet. The active development of the rust was not noticed until early March. But the unusually high rainfall in March led to a severe outbreak. The destructiveness, however, was somewhat checked by the dryness in April, when the relative atmospheric humidity averaged 66.2 per cent. At the end of April and in May, there was again unusually high rainfall, which in combination with a relatively low temperature delayed the maturity of wheat even more than in 1939. The activity of stripe rust was resumed for a while in early May. This year was characterized by the prolonged period of rust development as well as by the highest yields for most varieties of wheat.

The very dry winter of 1941, an intermediate rust year, appeared to be responsible for the late outbreak of stripe rust, which did not start until early March, and probably was induced by considerable rainfall in late February. The progress of rust, however, was somewhat checked by dryness in March and the teliospores soon formed. Toward the end of March the recurrence of rains allowed the rust to become prevalent for a rather short period. This year was even more unfavorable to wheat growth than to the development of rust. The dryness in spring and the high temperatures in April caused a precocious growth and very poor yield in wheat.

From December of 1941 to March of 1942 marks a period of drought, but the relative atmospheric humidity remained above 80 per cent. The year 1942 was an intermediate rust year. The rust started to gain in intensity only late in March. The unusually high rainfall in April, however, caused a sufficient retention of water both in the soil and in the plants to favor the development of the rust. This year was approximately intermediate among the seven years recorded in the prevalence of stripe rust, which was less destructive than in 1938, 1939, and 1940, but more destructive than in 1941, 1943, and 1944.

Both 1943 and 1944 were light rust years, the former being unfavorable to the growth of both wheat and stripe rust. Drought throughout the spring and winter kept down the rust, which was first noticed during mid-April. The rust situation of 1944 was comparable to that of 1943, though the dryness of the season was not so pronounced. The precipitation for 1944 was only slightly less than that for 1941 (Table 3), but its distribution varied. There was considerable rainfall in February, 1941, but much less in the same month of 1944. In both years March was dry, therefore the amount of precipitation in February would be important in determining the increase of actual inoculum early enough to initiate an epidemic at the right time.

From the foregoing discussions, it appears that the rainfall during the late winter and the spring is most important of all in affecting the prevalence of stripe rust in this region. However, not merely the amount of rainfall but its distribution as well should be taken into consideration. Rain should occur early in the season in order to initiate a rust epidemic. Besides its effect on the initiation of the disease, rainfall may render the wheat plants more susceptible to the rust. Both atmospheric humidity and cloudiness are effective in regulating the evaporation of water from plants as well as from soil. Low temperatures toward late spring favor repetition of the uredial stage and postpone the production of teliospores. These factors altogether have a profound influence on the increase of inoculum as well as the development of successive crops of urediospores during the growing season.

SUMMARY

In Chengtu plain, China, wheat rusts occur in the following sequence: (1) *Puccinia triticina*, (2) *P. glumarum*, (3) *P. triticina*, (4) *P. graminis*. The stripe rust caused by *P. glumarum*, however, is often the most important factor in reducing the yield of wheat.

Besides attacking wheat and barley, stripe rust occurs very commonly on *Agropyron ciliare* and *A. semicostatum* in the plain and surrounding mountains. On the grass hosts the rust appears later in the season than on wheat. Both the physiologic races isolated from *Agropyron* are different from those on wheat. Hence the grasses are not considered to have any importance in the epidemics of stripe rust on wheat.

The urediospores of stripe rust germinate best at 11.5° C. and the maximum lies around 25° C. Above 20° C. germination is much retarded and the final percentage is very low.

Under local conditions with high atmospheric humidity prevailing, the urediospores remain viable no longer than one month. The infectivity of spores often is retained a much shorter time, usually not more than one week. At high temperatures, the urediospores rapidly perish. At 36° C., the spores are viable for only two days.

In the plain the winter is so mild that stripe rust appears to have no difficulty in overwintering by means of either the urediospores or mycelium. Between crops of wheat, however, there exists a gap of five wet and hot summer months during which the stripe rust can hardly survive. On the mountains west of the plain, the season for winter wheat is much prolonged and spring wheat is cultivated at altitudes around 3,000 m. Stripe rust has been seen at high altitudes persisting on wheat throughout the summer. From those high levels, the urediospores are conveyed by wind southeastward into the lower altitudes to infect the fall sown wheat. The sources of inoculum for subsequent infections on wheat at higher altitudes are either the urediospores which have overwintered *in situ* or the urediospores blown back from the lower levels, or both.

The influence of climatic factors on the prevalence of stripe rust have been investigated from 1938 to 1944 at Chengtu. The results indicate that the amount and distribution of rainfall in late winter and spring are most important in determining the development of rust epidemics. Atmospheric humidity and cloudiness also are of significance because of their effects on the evaporation of water from the plants and from soil. Low temperatures in late spring delay the maturity of wheat and favor the repetition of the uredial stage of the rust. Among the seven years recorded, the worst stripe rust epidemic occurred in 1939, while there was very little rust in 1943 and in 1944. The other years are classed as intermediate.

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LITERATURE CITED

1. BECKER, J. Untersuchungen über die Lebenfähigkeit von Uredosporen von *Puccinia glumarum*. Kühn-Archiv **19**: 353–411. 1928.
2. BECKER, HANNA, and HELEN HART. Das Auftreten und die Verbreitung von Gelbrost im Ostharz und den daran angrenzenden Weizenanbaugebieten. Ztschr. Pflanzenkrank. u. Pflanzenschutz. **49**: 449–481. 1939.
3. BUTLER, E. J., and J. M. HAYMAN. Indian wheat rusts. Mem. Rept. Agr. India, Bot. Ser. **1**: 21–53. 1906.

4. DUCOMET, V. Quelques observations et expériences sur les rouilles des céréales. Rev. Path. Veg. et Ent. Agric. **12**: 124–128. 1925.
5. ERIKSSON, J. Ueber die Förderung der Pilzsporenkeimung durch Kälte. Centr. Bakt. II, **2**: 557–565. 1895.
6. HUNGERFORD, C. W. Studies on the life history of stripe rust, *Puccinia glumarum* (Schm.) Erikss. et Henn. Jour. Agr. Res. [U.S.] **24**: 607–620. 1923.
7. MEHTA, K. C. Observations and experiments on cereal rusts in the neighbourhood of Cambridge, with special reference to their annual recurrence. Trans. Brit. Myc. Soc. **8**: 142–176. 1923.
8. ————. The annual recurrence of rusts on wheat in India. 16th Indian Sci. Cong. (Sect. Bot.), Madras, 1929. 25 pp. 1929.
9. ————. Annual outbreaks of rusts on wheat and barley in the plains of India. Indian Jour. Agr. Sci. **1**: 297–301. 1931.
10. ————. The cereal-rust problem in India. Indian Jour. Agr. Sci. **1**: 302–304. 1931.
11. MENACACCI, M. Alcune ricerche sulle ruggini del frumento in agro Romano. Boll. R. Sta. Pat. Veg. **9**: 305–320. 1929.
12. NEWTON, M., and T. JOHNSON. Stripe rust, *Puccinia glumarum*, in Canada. Can. Jour. Res. **14**: 89–108. 1936.
13. RAEDER, J. M., and W. M. BEVER. Spore germination of *Puccinia glumarum* with notes on related species. Phytopath. **21**: 767–789. 1931.
14. STRAIB, W. Untersuchungen über das Vorkommen physiologischer Rassen des Gelbrostes (*Puccinia glumarum*) in den Jahren 1935/36 und über die Aggressivität einiger neuer Formen auf Getreide und Gräsern. Arb. Biol. Reichsanst. Land. u. Forstwirtschaft. **22**: 91–119. 1937.
15. STROEDE, W. Ueber den Einfluss von Temperatur und Licht auf Keimung der Uredosporen von *Puccinia glumarum* f. sp. *tritici* (Schmidt) Erikss. et Henn. Phytopath. Ztschr. **5**: 613–624. 1933.
16. WILSON, E. H. A naturalist in Western China. Vols. I and II. London. 1913.

COPPER SULPHATE AS AN ERADICANT SPRAY FOR POWDERY MILDEWS

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Powdery mildews are among the most easily controlled of fungus diseases of foliage. While a great variety of chemicals in liquid and dust form have been successfully used to control powdery mildews, sulphur dust was one of the first, and to this day remains the most universally successful. Liquid sprays containing lime-sulphur or wettable sulphur are, in general, similar in effectiveness to sulphur dust, but are more adhesive and more expensive to apply. Lime-sulphur spray is generally more injurious to the host and more effective as a fungus eradicant than either wettable sulphur or sulphur dust. For the relatively few cases where sulphur sprays or dusts are undesirable, one of several copper-containing sprays is usually used, and the writer feels that bluestone (copper sulphate, $CuSO_4 \cdot 5H_2O$) with a spreader is one of the most efficient, perhaps the most, reported to date.

Radclyffe (6) in 1861 was perhaps the first to use bluestone as a foliage spray. He used 2 oz. bluestone to a stable bucket of water (probably between 0.3 and 0.6 per cent) and poured it on the rose bushes from a fine spout. He reported that the roses were cleaned of powdery mildew. He recorded no foliage injury but his statement that he would use a lower dosage next year indicated that injury may have occurred. In commenting on Radclyffe's method of mildew control, another writer (1) cautioned against the use of bluestone on foliage, but he reported no experiments or observations. Two years later Radclyffe (7) repeated his recommendation of bluestone for control of rose mildew, though he also gave sulphur as an acceptable treatment.

Following the discovery of Bordeaux mixture, Millardet (4) in France made extensive tests of bluestone sprays on foliage for the control of grape downy mildew, and Taft (8, 9) in Michigan recommended 1–1000 to 1–4000 bluestone sprays on fruit trees in foliage. These treatments have disappeared from present recommendations. Blake (2) used bluestone spray at the rate of "about a tablespoon to a bucket of water," and claimed perfect eradication of rose mildew. In spite of these favorable though fragmentary reports, and in the apparent absence of any definite contrary evidence, the use of bluestone spray was apparently not accepted by the public or by plant pathologists, and the writer is aware of no recommendation of the use of bluestone for the control of a powdery mildew at present. Soluble coppers such as copper ammonia mixtures and copper oleate in oil, and less soluble coppers such as Burgundy mixture, Bordeaux mixture, and cuprous oxide are being successfully used for the control of powdery mildews, but little quantitative information on their comparative value is available.

The experimental demonstration that the eradicant value of Bordeaux for bean powdery mildew decreased with increasing amounts of lime in the

spray (14) stimulated this further study of bluestone as an eradicant spray for powdery mildew.

EFFECT OF A SPREADER ON THE SPRAY DEPOSIT ON FOLIAGE

Spray deposit was calculated from the measured wet deposit and the known fungicide content of the spray. If it can be assumed there is no selective adherence and that there is no physical change in the liquid induced by the suspended or dissolved fungicide, the deposit of any dosage of spray mixture could be calculated from the measured deposit of a similar appli-

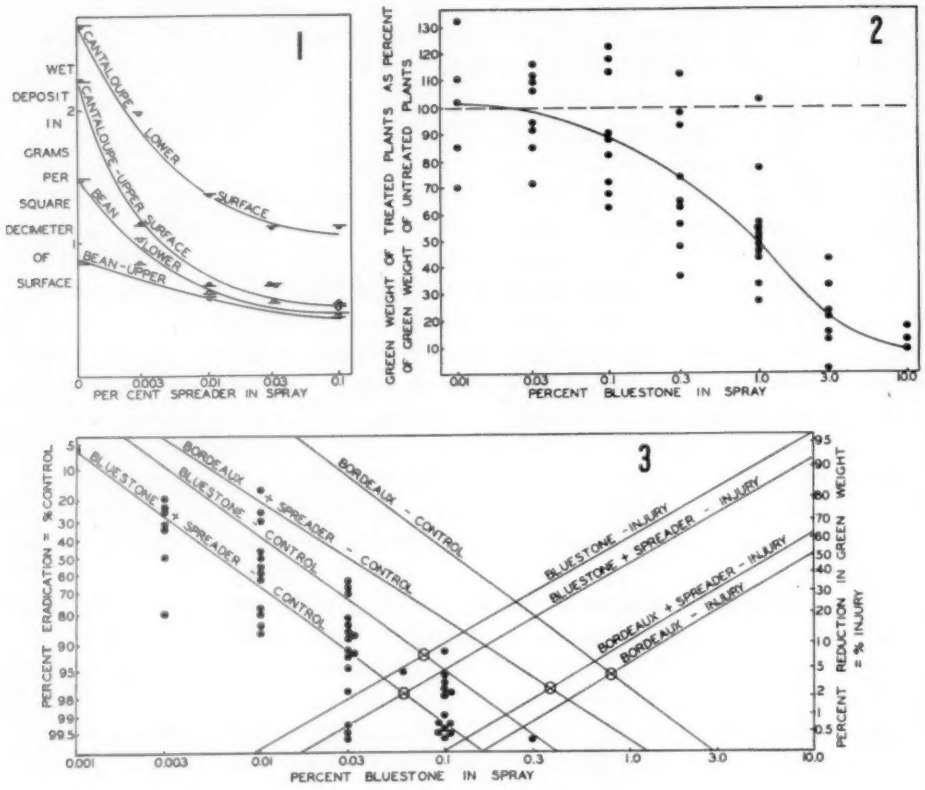

1
WET DEPOSIT IN GRAMS PER SQUARE DECIMETER OF SURFACE
CANTALOUPE - LOWER SURFACE
CANTALOUPE - UPPER SURFACE
BEAN - LOWER
BEAN - UPPER
PER CENT SPREADER IN SPRAY
2
GREEN WEIGHT OF TREATED PLANTS AS PERCENT OF GREEN WEIGHT OF UNTREATED PLANTS
PERCENT BLUESTONE IN SPRAY
3
PERCENT ERADICATION = % CONTROL
BORDEAUX + SPREADER - CONTROL
BLUESTONE + SPREADER - CONTROL
BORDEAUX - CONTROL
BLUESTONE - CONTROL
BLUESTONE - INJURY
BLUESTONE + SPREADER - INJURY
BORDEAUX + SPREADER - INJURY
BORDEAUX - INJURY
PERCENT REDUCTION IN GREEN WEIGHT = % INJURY
PERCENT BLUESTONE IN SPRAY

FIG. 1. The relation of concentration of spreader (glyceryl alkyl resin) in the spray to the wet deposit on the upper and lower surfaces of cantaloupe and bean leaves.

FIG. 2. The relation of concentration of bluestone in spray (with 0.05 per cent spreader in all cases) to injury to beans. Each dot represents the green weight of the leaves of young Pinto beans, grown in 4-inch pots in the greenhouse, 5 to 10 days after spraying with bluestone of the indicated concentration, expressed as a percentage of the green weight of similar unsprayed plants. The smooth curve approximates the average results.

FIG. 3. The relation of concentration of bluestone in spray to eradication of bean mildew and to injury of noninfected plants. Only the readings (dots) for bluestone plus spreader used as an eradicant are given. In all cases the straight line best fitting the average results is indicated. The Bordeaux mixture was prepared from equal parts of bluestone ($CuSO_4 \cdot 5H_2O$) and line (CaO), and the dosage is the percentage of bluestone in the final spray.

cation of water. While these assumptions are not justified under all conditions, their acceptance is believed to introduce no errors of consequence in the present study and data for water only will be presented here, though deposit data have been collected for lime-sulphur, Bordeaux, and Burgundy sprays. Weighed single leaves held in a vertical position were sprayed with a DeVilbiss atomizer with 35 lb. air pressure at a distance of about 10 inches until maximum deposit appeared to have been reached and runoff had just started and the leaf was again weighed. The leaf area was measured with a planimeter and the spray deposit was calculated as grams of wet spray per square decimeter of leaf surface. Glyceryl alkyl resin (B1956 spreader of Rohm and Haas Co.) was used as a spreader and the quantitative effect of spreader on deposit was determined. Average results of one test in triplicate are in figure 1. Water alone gave a deposit of 2.6 g. per sq. dcm. on the lower surfaces of cantaloupe leaves, which were poorly wetted but retained the water in large drops before runoff. Addition of 0.003, 0.01, and 0.03 per cent spreader improved the wetting of the leaves and reduced the deposit. With 0.03 per cent spreader there was apparently perfect wetting and a deposit of 1.2 g./dcm.^{2}; and further addition of spreader had no apparent effect. Similar additions of spreader had similar relative effects on the deposit on the upper surfaces of cantaloupe leaves and on the upper and lower surfaces of bean leaves, though the actual deposit was lower on these surfaces. A concentration of 0.05 per cent glyceryl alkyl resin was therefore arbitrarily chosen as the dosage to be used in tests where the addition of spreader was involved, and spraying was always continued until runoff had begun.

BLUESTONE INJURY TO FOLIAGE

Spray injury was quantitatively determined by spraying healthy plants with a series of dosages of the test spray and weighing the foliage after maximum injury had developed. This was usually 5–10 days after spraying. The reduction in green weight was a measure of spray injury. Detailed results of spray injury to beans from bluestone plus spreader in 11 tests in the greenhouse are in figure 2 and the data show considerable variation, but significant injury did not result until concentrations of 0.1 per cent bluestone and above were used. For convenient comparison with the data on eradication of bean powdery mildew, the average data on injury to bean foliage from bluestone with and without spreader and from Bordeaux with and without spreader are expressed in figure 3 as the nearest straight line on a logarithmic probability scale (10). To produce 5 per cent reduction in green weight required a spray dosage of 0.062 per cent bluestone without spreader, 0.11 per cent bluestone with spreader, 0.27 per cent Bordeaux with spreader, and 0.77 per cent Bordeaux without spreader. The greater injury from bluestone without spreader than from bluestone with spreader is probably due to the greater deposit without spreader (See figure 1), but the cause of the greater injury from Bordeaux with spreader than from Bordeaux without spreader is undetermined.

Plant injury was also estimated in some tests on a scale of 0 to 100 in which 0 indicated no injury and 100 indicated death of all leaves. Data from all tests of host injury with bluestone sprays with and without spreader are in table 1. With beans it was found that the rating and weighing methods gave similar results. Spray concentration for 50 per cent injury ranged from a minimum of 0.035 per cent bluestone without spreader for greenhouse-grown mustard to over 10 per cent bluestone for garden beet in the greenhouse. With most plants the addition of a spreader increased the dosage necessary for 50 per cent injury. With peas in the greenhouse, however, bluestone spray with spreader caused more injury than bluestone spray without spreader. This probably was because practically no wetting of pea

TABLE 1.—*Foliage injury from bluestone spray*

Plant	Location	No. of tests	Method of estimating injury	Dosage (per cent bluestone in spray) for 50 per cent injury	
				Without spreader	With 0.05 per cent spreader[a]
Bean	Greenhouse	11	weight	0.66	1.2
do	do	3	rating	0.5	1
do	Field	1	do	0.8	1.8
Cucumber	Greenhouse	6	weight	0.5	1.0
do	Field	1	rating	0.3	0.8
Cantaloupe	do	1	do	0.3	0.8
Gardenbeet	Greenhouse	1	do	> 10	> 10
Grape	Field	1	do	3	4
Pea	Greenhouse	2	weight	10	3
do	Field	1	rating	1	2
Hop	do	1	do	1.8	1.8
Mustard	Greenhouse	2	weight	0.035	0.15
Potato	do	1	rating	.....	1.5
Tomato	do	2	do	.....	1.5
Rose	Field	1	do	1	1
Apple	do	1	do	.....	> 0.3

[a] Glyceryl alkyl resin.

leaves occurred unless a spreader was added. With increasing amounts of spreader (0.05, 0.15, and 0.45 per cent) added to bluestone solution, injury to peas in the greenhouse was progressively increased. With peas in the field, however, where the foliage was less difficult to wet, injury was apparently greater without a spreader than with one. Foliage injury from bluestone sprays is greater when the spray dries slowly than when it dries rapidly, is greater from applications to the lower leaf surface than from applications to the upper leaf surface, and is greater on mildew-infected than on healthy plants. Many other factors are probably important in determining the severity of leaf injury from bluestone. On one occasion (Dec. 31, 1944) greater injury to greenhouse beans resulted from an application of 1 per cent Bordeaux than from 1 per cent bluestone, and in several tests greater injury has resulted from Bordeaux without spreader than from Bordeaux with spreader.

ERADICATION OF POWDERY MILDEWS

The infected plants were sprayed with the test fungicide, and the amount of living mildew on check and treated plants was estimated 5 to 10 days after spraying on an arbitrary rating scale of 0 to 10 in which 0 indicated no mildew present and 10 that the leaf surface was entirely covered. The mildew present before spraying, but killed by the spray treatment, was usually still apparent on the leaves at time of recording the incidence of mildew, but was easily distinguished from the living mildew and was not included in the record. In some tests the mildew colonies were counted on unit areas and the counting method gave results parallel to the rating method; but a lower dosage of inoculum was necessary for the counting method because with heavy inoculation the powdery mildew colonies merged and could not be individually distinguished with the unaided eye by the time infection became evident. Upper and lower leaf surfaces were rated separately. Control was calculated by expressing the difference in rating between treated and check plants as a percentage of the rating for the checks.

Average results, presented as the best straight line fitting the data on a logarithmic probability scale (10) for greenhouse tests of the eradication of bean powdery mildew where a range of concentrations of bluestone and Bordeaux with and without spreader was applied 4 to 8 days after inoculation are in figure 3 and the detailed values for bluestone plus spreader are indicated. In this figure are also given the data on injury to healthy beans by these same sprays. Since the four curves for eradication are apparently approximately parallel, and the four curves for injury are also approximately parallel, all materials can be conveniently compared for eradication and injury; and the intersection points for eradication and injury by the same materials, indicated as circles, offer a good basis of overall comparison of the four sprays studied here. Bluestone plus spreader required a spray dosage of only 0.042 per cent bluestone for 95 per cent control by eradication while bluestone without spreader, Bordeaux plus spreader, and Bordeaux without spreader required about 2.5, 6, and 18 times this dosage, respectively, for equivalent control under these conditions. It is, of course, the balance between plant injury and disease incidence (as affected by eradication in this case) which is the best index of merit of a material, and which determines the better of a group of fungicides. For results as presented in figure 3, the lower the level of the intersection point of the control and injury curves, the lower the total of disease plus injury at that and similar dosages; and if low percentage disease and low percentage injury are of equal quantitative merit, the optimum dosage will be indicated by the point of intersection of the control and injury curves, disregarding such factors as cost of materials. On this basis as well as on the basis of the lowest dosage for control, bluestone plus spreader was the most effective of the four sprays compared. The optimum concentration of bluestone plus spreader was 0.06 per cent bluestone which was associated with 2 per cent disease and 2 per cent injury, the optimum concentration of bluestone without spreader was

0.08 per cent bluestone which was associated with 8 per cent disease and 8 per cent injury, the optimum concentration of Bordeaux plus spreader was 0.4 per cent Bordeaux which was associated with 3 per cent disease and 3 per cent injury, and the optimum concentration of Bordeaux without spreader was 0.8 per cent Bordeaux which was associated with 4 per cent disease and 4 per cent injury. Increasing the dosages above these would decrease the disease (or increase the control by eradication) but the increase in injury would be greater than the decrease in disease. For example, 0.1 per cent bluestone with spreader would give about 0.7 per cent disease and 5 per cent injury, or a total of 5.7 per cent disease plus injury, while at the optimum dosage of 0.06 per cent bluestone plus spreader the disease plus injury was only about 4.8 per cent. The specified values are not sufficiently significant that differences of such small magnitude due to spray dosage can be expressed with any degree of certainty, but the principle would appear valid for this or smaller or greater differences.

Fewer and less detailed studies of bluestone plus spreader as an eradicant spray have been made on the powdery mildews of cucumber, cantaloupe, mustard, grape, rose, peach, and apple, and in all cases approximately perfect eradication of powdery mildews was secured with approximately 0.1 per cent bluestone plus spreader.

With rose powdery mildew, plants of the varieties Night, Crimson Glory, and Rose Picture, grown 8 feet apart in a field plot at Albany, California, were sprayed or dusted May 10, May 29, June 21, and July 12, 1944, with 0.01 per cent bluestone plus spreader, 0.03 per cent bluestone plus spreader, 0.1 per cent bluestone plus spreader, 1 per cent lime-sulphur plus spreader, and sulphur dust, in a non-replicated series, and 4 checks of each variety were maintained throughout the plot. Mildew severity was rated May 15, July 3, and July 17, and 99.8, 100, 100, 97.3, and 100 per cent control, respectively, resulted from these treatments. Mildew was present, but not severe, on all check plants. There was no plant injury apparent from sulphur dust or 0.01 per cent bluestone, but 0.03 per cent bluestone caused slight injury noticeable on Rose Picture only, while 0.1 per cent bluestone caused injury estimated at about 10 per cent on all varieties. Even this amount of injury could easily have been overlooked.

OTHER FORMS OF COPPER AS ERADICANT SPRAYS

Sprays containing the soluble coppers, copper sulphate, copper nitrate, copper chloride, copper acetate, and copper-ammonia mixtures have been compared with sprays containing the relatively insoluble coppers, Bordeaux, Burgundy, cuprous oxide, copper phosphate, copper carbonate, and copper arsenite for the eradication of bean powdery mildew in the greenhouse. In general, the soluble coppers were eradicant at lower dosages than the insoluble coppers, and copper nitrate, copper chloride, and copper acetate were similar in effectiveness to copper sulphate. Burgundy mixtures containing one and one-half parts of sodium carbonate for each part of bluestone were

effective at lower dosages than the same concentrations of copper as Bordeaux containing equal amounts of bluestone and lime. With varying bluestone : lime ratios the eradicant value of Bordeaux decreased as the amount of lime in the spray increased.

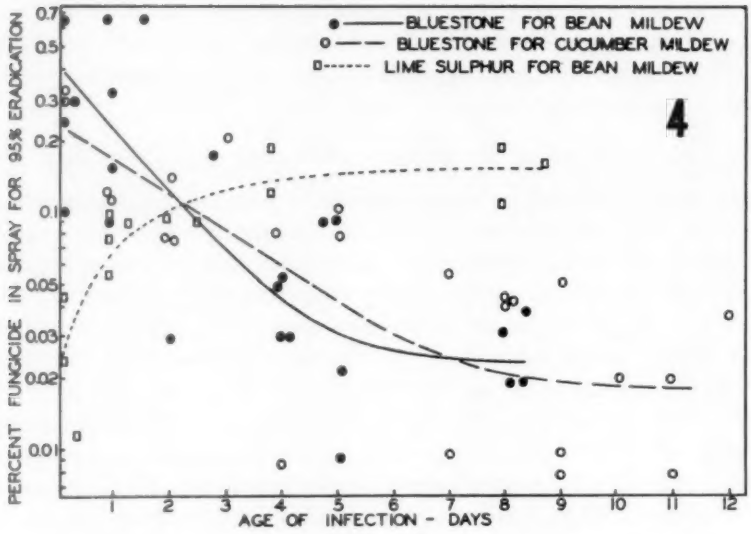
NON-TOXICITY OF BLUESTONE TO POWDERY MILDEW CONIDIA

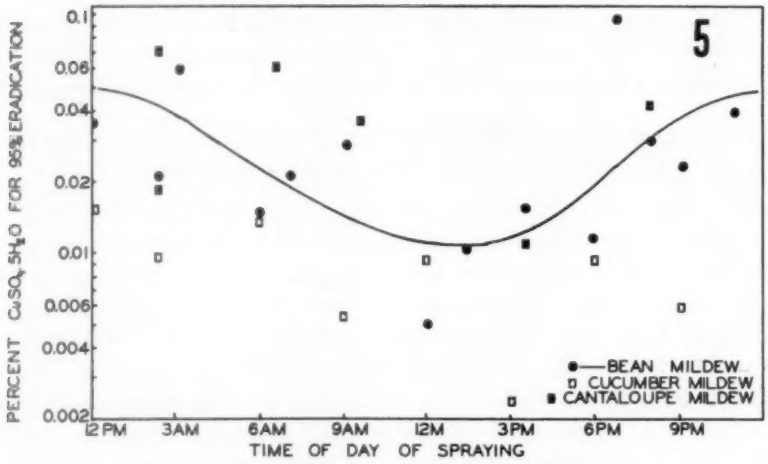
Conidia of *Erysiphe polygoni* from bean and from mustard dusted on the surface of 10 per cent copper sulphate solution germinated to the extent of 17 and 28 per cent, respectively, when controls on water germinated to the extent of 28 and 46 per cent, respectively, in the average of two tests. The conidia remained on the surface of the liquid and were not readily wetted and the germ tubes frequently came in contact with the bluestone solution without apparent injury, except that they were somewhat shorter than on water. When wetting agents were added to bluestone solutions of similar or lower strength, germination of conidia of bean mildew was prevented, but the wetting agent alone in water inhibited germination, and it was not determined to what extent the inhibition of germination was due to bluestone or to spreader. However, in view of these results the writer feels that with powdery mildews, as with downy mildews (12) and rusts (14), tests of the effect of chemicals on the germination of spores on glass are of relatively little value in appraising chemicals for the control of these diseases.

EFFECT OF AGE OF INFECTION ON THE SUSCEPTIBILITY OF POWDERY MILDEWS TO ERADICATION BY BLUESTONE SPRAY

Age of infection is defined for purposes of this paper as time from inoculation. Beans and cucumbers artificially inoculated with their respective powdery mildews were sprayed with a range of concentrations of bluestone plus spreader at various intervals. For each concentration series the concentration of bluestone for 95 per cent eradication was determined by plotting the observed results on a logarithmic probability scale (10). The LD95 (dosage for 95 per cent control) for different ages of infection, as determined in 5 tests with bean powdery mildew and in 3 tests with cucumber powdery mildew (Fig. 4), decreased from about 0.3 per cent bluestone with spreader at 2 hours (0 days) to about 0.03 per cent at 8 days from inoculation. This increasing sensitivity of powdery mildews to bluestone as the age of infection is increased is the opposite of what was expected, but might be considered in accord with the results on spore germination *in vitro* already presented, since it has been shown that bluestone was relatively non-toxic to germinating conidia.

Results of three tests of the effect of age of infection on the LD95 for lime-sulphur spray performed concurrently with the bluestone are also presented in figure 4. According to these data bean powdery mildew becomes increasingly resistant to lime-sulphur as the infections become older, or the tendency is the opposite to that with bluestone. The results with lime-sulphur cannot be considered conclusive, however, since this tendency was not





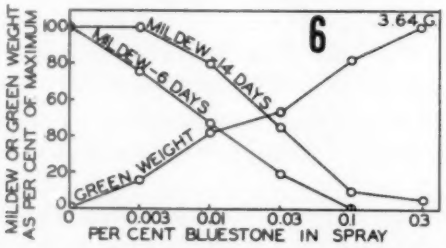


FIG. 4. The relation of age of infection (days after inoculation) to the dosage required for 95 per cent eradication of bean and cucumber powdery mildews with bluestone spray plus spreader, and for bean powdery mildew with lime-sulphur spray plus spreader.

FIG. 5. The relation of time of day of spraying to the dosage of bluestone spray (with 0.05 per cent spreader) required for 95 per cent eradication of bean powdery mildew, cucumber powdery mildew, and cantaloupe powdery mildew.

FIG. 6. The relation of concentration of bluestone spray (with 0.05 per cent spreader) to eradication of bean powdery mildew and to the green weight of the same plants.

corroborated in two other tests, not run concurrently with the tests of bluestone.

EFFECT OF TIME OF DAY WHEN SPRAYS ARE APPLIED

Results of tests to determine the effect of time of day of spraying on the eradicant value of bluestone plus spreader applied 4 to 8 days after inoculation in 3 greenhouse tests with bean powdery mildew, 2 tests with cucumber powdery mildew, and 3 tests with cantaloupe powdery mildew are in figure 5. The average LD95 for bluestone with spreader for bean powdery mildew, presented as a smooth curve, ranged from a maximum of 0.05 per cent bluestone at 12 p.m. to a minimum of about 0.01 per cent at 12 noon. The limited number of tests, the variability of the data, and the apparently higher LD95 values secured in the tests summarized in figure 3 should be emphasized in considering the reliability of this apparent diurnal variation in the susceptibility of bean mildew to eradication by bluestone. In 2 tests each with cucumber powdery mildew and cantaloupe powdery mildew, tendency toward diurnal difference was not clearly apparent.

LOW PROTECTIVE ACTION OF COPPER SPRAYS

Most copper sprays or dusts tested to date have had little protective action against the powdery mildews studied, while sulphur sprays and dusts have had marked protective as well as eradicant action. Evidence of the greater eradicant than protective action of copper sprays and even of the stimulation of bean powdery mildew by protective applications of Bordeaux mixture has already been presented (13), but further data on protection derived from copper sprays will be presented here.

Results secured in protection spray tests with powdery mildews depend on the method of inoculation to a much greater extent than do results in eradication tests. The method of inoculation adopted as standard for tests in the greenhouse has been spraying of the test plants in the afternoon with a water suspension of powdery mildew conidia. On plants that had not received protective applications of fungicides, this method of inoculation has yielded heavier and more uniform infection of upper and lower leaf surfaces than any other method, though inoculation by dusting conidia direct from infected to healthy plants has usually resulted in heavier infection on the upper leaf surface, especially on plants treated with a protective fungicide. Powdery mildew conidia are normally and successfully disseminated in nature by being blown by air currents from a dry infected surface to a dry noninfected surface, therefore, inoculation by spraying plants with a spore suspension is unnatural. Furthermore, water is directly injurious to powdery mildews (11), especially at high impact pressures, and copper fungicides may be more toxic to powdery mildews in the presence of free water than in its absence (13).

Despite the admitted objections to the method, most studies of protective and eradicant sprays for powdery mildews have involved spraying with a spore suspension as a method of inoculation. In 6 closely comparable tests,

the LD95 for bluestone plus spreader against bean powdery mildew was about 3 per cent bluestone for protection (determined by extrapolation, since bluestone caused so severe host injury that protection was difficult to determine at dosages above one per cent) and about 0.03 per cent bluestone for eradication; or the dosage for equivalent control by eradication was only about one one-hundredth that required for protection. In similar tests the LD95 for Bordeaux plus spreader was about 1.5 per cent for protection, and 0.4 per cent for eradication. Bluestone without spreader, Bordeaux without spreader, Burgundy mixture, cuprous oxide, and lime-sulphur have also been better as eradicant than as protective sprays though in no case was the difference as great as with bluestone. Emulsified cottonseed oil (tested in 2 paired comparisons at 3 dosages only) is the only spray tested which has given greater control by protection than by eradication. Tests of the same materials for cucumber powdery mildew indicate that with this disease also sprays are usually more effective as eradicants than as protectives.

When inoculation has been by dusting the sprayed plants with dry powdery mildew conidia, control by protective sprays has been even less than when inoculation was by spraying with a spore suspension.

INCREASE IN GREEN WEIGHT AND YIELD OF SPRAYED PLANTS

The relation of dosage of bluestone spray to control of powdery mildew and to green weight and yield of plants has been measured in greenhouse and field tests with bean powdery mildew, cucumber powdery mildew, and cantaloupe powdery mildew. Results of some representative tests are given in figures 6–9. In the test represented in figure 6, duplicate 4-inch pots of two bean plants each were inoculated with powdery mildew on January 7, ten days from seeding on December 28.

On January 14, 7 days from inoculation, the growing point of each bean plant, that is, the growth beyond the inoculated primary true leaves, was removed, and the plants were sprayed with a series of concentrations of bluestone plus 0.05 per cent spreader. On January 20, 6 days after spraying and 14 days after inoculation, the severity of mildew on the upper and lower surfaces of the leaves was rated, and the average results are in figure 6 as the curve "mildew—6 days." Eight days later (mildew—14 days) mildew had increased considerably on all sprayed plants. The mildew on the plants treated with 0.1 and 0.3 per cent bluestone was probably due to reinfection after spraying. On January 31, 17 days after spraying, the new growth (axillary shoots) which had formed in the axils of the two primary true leaves since spraying was cut off and weighed, and it is indicated in figure 6 as green weight. This green weight varied in a fairly straight line relation from 0 grams on the similarly decapitated check plants to 3.64 grams on the plants sprayed with 0.3 per cent bluestone. At this time the primary leaves of the check plants and those sprayed with 0.003 per cent bluestone, all heavily infected, were dead.

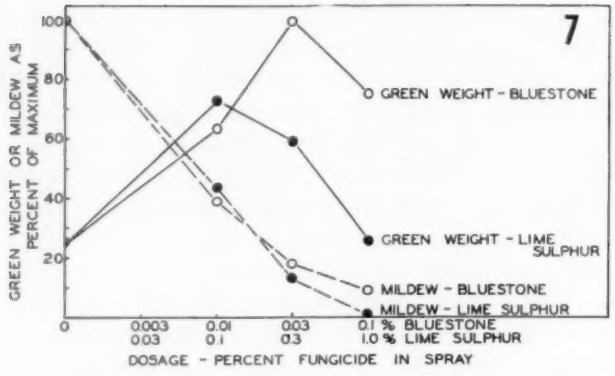
In another similar test, beans seeded in the greenhouse, February 10, and inoculated February 21, were shaken occasionally to secure a fairly heavy

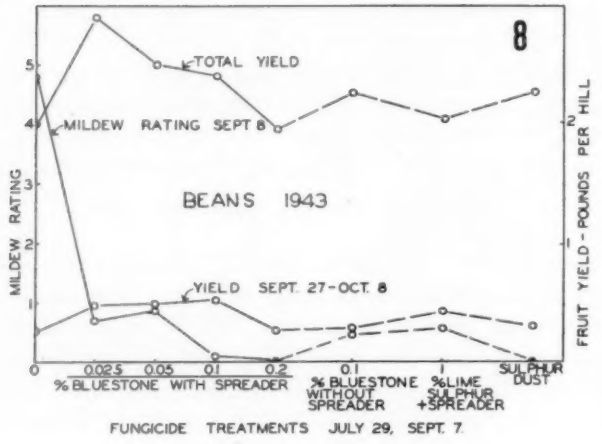
reinfection of the new growth. On February 29 the growing points of the plants were removed and the plants, in duplicate pots, were sprayed on February 29, March 6, March 26, and April 7 with spreader only, 0.001 per cent bluestone plus spreader, 0.003 per cent bluestone plus spreader, 0.01 per cent bluestone plus spreader, 0.03 per cent bluestone plus spreader, 0.1 per cent bluestone plus spreader, and 0.3 per cent bluestone plus spreader. The controls were unsprayed. On April 19, all leaves on the unsprayed plants were dead and the green weight of leaves plus fruit of the sprayed plants increased from 5.9 g. per plant for the plants sprayed with spreader only to 31.5 g. for plants sprayed with 0.03 per cent bluestone, but was only 15.5 g. for plants sprayed with 0.3 per cent bluestone. Records of mildew infection showed a progressive reduction in mildew with increasing concentration up to 0.1 per cent bluestone, which gave perfect eradication under these conditions.

The procedure of removing the growing point of each plant soon after inoculation, as in the two tests just reported, usually results in differences in green weight yield due to mildew infection and due to control of mildew which are greater and appear sooner than in non-decapitated plants, though differences of similar direction but lower magnitude resulted from the use of non-decapitated plants.

While bean powdery mildew apparently can be easily and thoroughly controlled by the use of bluestone spray in the greenhouse and in the field, this spray treatment gives no better control and is more laborious to apply than the standard dusting with sulphur. Bean powdery mildew lends itself more easily to intensive greenhouse study than any other powdery mildew known to the writer, and it is believed that results secured with this disease will apply in large part to many other powdery mildews, some of which cannot be commercially controlled with sulphur dust. Such a disease is cantaloupe powdery mildew, because cantaloupes may be severely injured by sulphur dust (5) as well as sulphur-containing sprays. Data of a comparative test of lime-sulphur and bluestone sprays for the control of cantaloupe powdery mildew in the greenhouse are in figure 7. Cantaloupes seeded in 4-inch pots December 20 were placed under conditions of moderately heavy infection from older infected plants, 6 replications were sprayed with each dosage of lime-sulphur and bluestone (plus 0.05 per cent spreader in all cases) on January 20, February 5, and February 25, and the green weight of the leaves was determined on March 11. For the dosages chosen, approximately equivalent mildew eradication was secured when bluestone was compared with lime-sulphur of about 10 times the dosage. With bluestone the green weight of the plants increased with increasing bluestone dosage and decreasing mildew up to 0.03 per cent bluestone, but then fell at 0.1 per cent bluestone. With lime-sulphur on the other hand, yield increased with 0.1 per cent lime-sulphur but was less with applications of 0.3 and 1 per cent lime-sulphur because of plant injury (Fig. 7).

In field tests, differences in mildew and in yield due to spray treatment have been less than in greenhouse tests. Kentucky Wonder beans seeded in


7
GREEN WEIGHT OR MILDEW AS PERCENT OF MAXIMUM
GREEN WEIGHT - BLUESTONE
GREEN WEIGHT - LIME SULPHUR
MILDEW - BLUESTONE
MILDEW - LIME SULPHUR
0.003 0.01 0.03 0.1 % BLUESTONE
0.03 0.1 0.3 1.0 % LIME SULPHUR
DOSAGE - PERCENT FUNGICIDE IN SPRAY



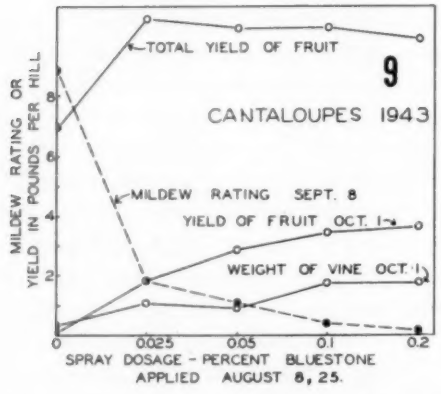

9
TOTAL YIELD OF FRUIT
CANTALOUPES 1943
MILDEW RATING SEPT. 8
YIELD OF FRUIT OCT. 1
WEIGHT OF VINE OCT. 1
MILDEW RATING OR YIELD IN POUNDS PER HILL
0.025 0.05 0.1 0.2
SPRAY DOSAGE - PERCENT BLUESTONE
APPLIED AUGUST 8, 25.

FIG. 7. The relation of concentration of bluestone spray plus spreader and of lime-sulphur spray (32° Baume) plus spreader to powdery mildew control and green weight in greenhouse test with cantaloupes.

FIG. 8. The relation of spray material to powdery mildew infection and yield of Kentucky Wonder beans in field plots.

FIG. 9. The relation of dosage of bluestone spray plus spreader to powdery mildew control and to yield of No. 45 cantaloupes in field plots.

hills May 6 were inoculated occasionally by dusting lightly with conidia from greenhouse plants and were sprayed in 7 replications on July 29 and September 7 with the materials indicated in figure 8. Mildew did not become severe, but marked control was secured with all treatments, though perfect control was secured only with 0.2 per cent bluestone plus spreader and with sulphur dust. Yields of green beans were recorded August 19, September 3, 13, and 27, and October 8. The highest total yield was from plants sprayed with 0.025 per cent bluestone plus spreader, but 0.025, 0.05, and 0.1 per cent bluestone all resulted in a higher yield than the check, and the relative increase was greatest in the last two pickings on September 27 and October 8.

With cantaloupe, previous tests had adequately demonstrated the danger of severe host injury from standard sulphur dusts and sprays and only bluestone (with 0.05 per cent spreader) was used in the 1943 tests. Cantaloupes seeded May 6 were artificially inoculated several times and were sprayed in 5 replicates of each treatment on August 8 and August 25. Mildew was severe and sprays containing 0.025 to 0.2 per cent bluestone gave marked control (see figure 9) which increased with dosage but was not perfect even with 0.2 per cent bluestone. Fruit yield was determined on September 3, 13, and 23, and October 1. Bluestone apparently delayed maturity and, on September 3 and 13, the fruit yield was greater on the check than on the treated plots (data not given in figure 9). The total yield, however, was greater for treated than for check plots. The most striking effect of spraying on yield was apparent at the final harvest on October 1, when the yield per hill ranged in a smooth curve from 0 for the control vines, which were mostly dead, to 3.52 lb. per hill for the vines treated with 0.2 per cent bluestone.

Field tests with cucumber powdery mildew in 1943 gave results similar to those presented for cantaloupe, except that the disease was not so severe and the yield increase from the treatments was less than in the case of cantaloupes.

DISCUSSION

Some explanation should be offered why a method of powdery mildew control reported in 1861 and neglected for 80 years should be revived in 1945. In the first place no one seems to have tried Radclyffe's bluestone treatment adequately and reported results, and Radclyffe's procedure was in part discredited by the warning of an anonymous writer as well as by the general knowledge of the injurious action of bluestone on foliage. Radclyffe's dosage, though not accurately stated, was much too strong. Also, his method of application was inefficient and his mixture lacked a spreader. The absence of a spreader in bluestone spray made it the poorest of the four spray mixtures compared in the present work (Fig. 3). Most important of all, perhaps, is the fact that sulphur dust for the control of powdery mildew was becoming well known in Radclyffe's time and, on plants tolerant to sulphur, no more efficient fungicide is yet known.

The suggestion in this paper that bluestone plus spreader is an effective practical control for several powdery mildews is limited to those cases where

sulphur dust or lime-sulphur are injurious to the host or give inadequate control. The best example of limitation of the use of sulphur because of host injury is cantaloupe powdery mildew (5), but there are undoubtedly others. No examples of failure to control powdery mildews with sulphur dust are known to the writer with certainty, but many have been indicated by observers. Such is indicated by Jacob (3) for established infections of grape powdery mildew and he recommends potassium permanganate plus sodium silicate and sodium bicarbonate. In tests by the writer, not reported here, bluestone plus spreader has proved a more effective eradicant for powdery mildews than the permanganate mixture. Many other spray materials, water alone, sea water, and solutions or suspensions of sodium carbonate, sodium bicarbonate, potassium hydroxide, nitric acid, sulphuric acid, sodium thiosulphate, ferrous sulphate, mercuric chloride, sodium silicate, zinc sulphate, vegetable, animal, and mineral oils, soaps, and materials used as spreaders have been tested, but none has been as effective at as low concentrations as bluestone plus spreader. Bluestone plus spreader has relatively little protective action, but effective practical control seems to be possible from its eradicant action alone.

SUMMARY

Addition of spreader to water sprays decreased the deposit but increased the coverage on cantaloupe and bean leaves. Wet spray deposit on lower leaf surfaces of cantaloupe decreased from about 2.6 g. per sq. dcm. when sprayed without spreader to 1.25 g. per sq. dcm. when spreader was added to the spray; and on lower leaf surfaces of bean the corresponding decrease was from 1.5 g. to 0.5 g. per sq. dcm.

Injury from bluestone sprays was measured on field- or greenhouse-grown bean, cucumber, cantaloupe, beet, grape, pea, hop, mustard, potato, tomato, rose, and apple foliage. The concentration of bluestone spray to cause 50 per cent injury varied from a minimum of 0.035 per cent bluestone for mustard to a maximum of over 10 per cent bluestone with beet. Bluestone injury was usually less when a spreader was added to the spray, but Bordeaux injury to greenhouse-grown beans was greater when a spreader was added. A 50 per cent injury to bean required about 10 times as much copper in the form of Bordeaux as in the form of bluestone.

Eradication of bean powdery mildew was secured at lowest copper dosages with bluestone plus spreader. For 95 per cent eradication a spray containing about 0.04 per cent bluestone plus spreader was required, while similar control required about 2.4 times as much copper in the form of bluestone without spreader, 6.4 times as much in the form of Bordeaux plus spreader, and 18 times as much in the form of Bordeaux without spreader. On the basis of maximum control with minimum injury, 0.06 per cent bluestone plus spreader was the most effective spray, followed in order by 0.4 per cent Bordeaux plus spreader, 0.8 per cent Bordeaux without spreader, and 0.08 per cent bluestone without spreader. The other soluble coppers (copper chloride, copper nitrate, and copper acetate) appeared to be about equal to

copper sulphate as eradicant sprays, but all insoluble coppers including Bordeaux, Burgundy, cuprous oxide, basic copper sulphate, copper oxychloride, and copper carbonate were distinctly less effective. Bluestone plus spreader was also superior to several other non-sulphur and non-copper chemicals tested.

Conidia of *Erysiphe polygoni* from bean and from mustard germinated well on the surface of solutions containing 10 per cent bluestone.

The spray concentration necessary for 95 per cent eradication of bean powdery mildew and cucumber powdery mildew decreased from a maximum of about 0.3 per cent bluestone applied at time of inoculation to about 0.03 per cent bluestone applied 8 days after inoculation. Eradication of bean mildew was secured from lower concentrations of bluestone spray applied during the day than from applications at night.

Heavier dosages of most sprays were necessary for protection than for eradication. For 95 per cent control about 100 times as much bluestone was necessary in a protective application as in an eradicant application.

The green weight of foliage and the yield of fruit on bean, cucumber, and cantaloupe plants on which powdery mildew was controlled with eradicant applications of bluestone plus spreader in greenhouse and field tests was greater than on similar unsprayed plants.

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LITERATURE CITED

1. ANONYMOUS. Rose mildew. Gard. Chron. 21: 1008. 1861.
2. BLAKE, JAMES. Mildew on roses—the bluestone remedy. The California Hort. and Floral Mag. 9: 353–354. 1879.
3. JACOB, H. E. Powdery mildew of the grape and its control in California. Cal. Agr. Ext. Serv. Cir. 31. 1929.
4. MILLARDET, M. Communication à la Societé d'Agriculture de la Gironde, 1 Avril, 1885. *In* Le cinquantenaire de la Bouille Bordelaise. Supplement à la Rev. Path. Veg. et Ent. Agr. de France 22: 22–27. 1935.
5. MILLER, P. A., and J. T. BARRETT. Cantaloupe powdery mildew in the Imperial Valley. Cal. Agr. Exp. Stat. Bull. 507. 1931.
6. RADCLYFFE, W. F. Rose mildew. Gard. Chron. 21: 967. 1861.
7. ————. Rose culture. Gard. Chron. 24: 701. 1864.
8. TAFT, L. R. Copper sulphate solution as a fungicide. Amer. Agric. 56: 34. 1895.
9. ————. When and what to spray. Mich. Agr. Exp. Stat. Special Spraying Bull. 1895.
10. WILCOXAN, FRANK, and S. E. A. MCCALLAN. Theoretical principles underlying laboratory toxicity tests of fungicides. Contr. Boyce Thomp. Inst. 10: 329–338. 1939.
11. YARWOOD, C. E. Control of powdery mildew with a water spray. Phytopath. 29: 288–290. 1939.
12. ————. Evaluating sulphur fungicides for the control of downy mildews. (Abstr.) Phytopath. 31: 865. 1941.
13. ————. Stimulatory and toxic effects of copper sprays on powdery mildews. Amer. Jour. Bot. 29: 132–135. 1942.
14. ————. The function of lime and host leaves in the action of Bordeaux mixture. Phytopath. 33: 1146–1156. 1943.

TIP BURN OF SUGAR BEET WITH SPECIAL REFERENCE TO SOME LIGHT AND NITROGEN RELATIONS

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Tip burn of sugar beet is a disease believed due to excessive accumulation of a substance or substances normally present in the plant. It has not been so diagnosed previously. Boncquet (1), in a paper concerned chiefly with curly top, described it incidentally and briefly under the designation "black edge" or "black tip." He at first thought it one of the types or stages of curly top, but later concluded that it was not. Robbins (2), in a study of sugar-beet mosaic, gave a somewhat more extensive description of tip burn which he regarded as an aspect of the mosaic.

DESCRIPTION OF SYMPTOMS

The symptoms of tip burn on vegetative beets in the field are distortion of the blades and death of more or less of the edge tissue often including the tip. The leaf blades may be cupped downward or in some plants the margins may be drawn in an upward curl. Frequently the leaf tissue between the large lateral veins becomes puffy and the blades then have a corrugated appearance. Much of the malformation of leaf blades appears to result from a greater retardation of growth of the vascular system than of the mesophyll tissue. The necrotic edges are tightly stretched. Often the tissue adjacent to the dark brown or black edges is yellow. These symptoms are usually on mature or nearly mature leaves. Occasionally, on plants where some of the larger leaves have these symptoms, the younger leaves may have severely truncated blades with blackened edges or even merely petioles with blackened tips (Fig. 1).

Affected beet plants with the characteristic downward cupping and necrosis on the edges of intermediate-aged leaves often have other symptoms that manifest early phases of tip burn (Fig. 2). Leaves younger than those with the conspicuous cupping may be blanched a yellowish green, with inconspicuous brown flecks, at the edges and tips. The marginal tissue so affected later dies and turns black. Development of these and other symptoms of tip burn seems to depend mainly on the duration of the conducive conditions.

Mother beets, especially when grown for seed in the greenhouse during winter, may have severe symptoms of types noted for vegetative beets and, in addition, the tips of the terminal or apical flowering shoot often blacken and die (Fig. 2).

Complete recovery is a regular characteristic of tip burn. The sub-

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sequently developed leaves in vegetative beets or leaves and axillary shoots in flowering beets are entirely normal (Fig. 3).

Tip burn has also been observed on Swiss chard.

Sugar beet tip burn must be distinguished from injury induced by an insect, *Lygus spp.* Potato tip burn (3) and hopper burn (4) present an analogous situation. Tips of Lygus-affected sugar-beet leaves wilt, die, and darken, and often a good deal of the adjacent tissue turns yellow. Such symptoms accompany dark swollen lesions or open scars on the midribs and petioles resulting from the feeding punctures. Usually also there is some distortion of the affected leaves associated with the bending of the midrib

FIG. 1. Sugar-beet leaves with some of the symptoms of tip burn. Leaves distorted with blanched constricted margins and early stages of edge and tip necrosis. Leaves collected from highly fertilized field planted in September near Riverside, California. Photographed December 29, 1943.

where severely injured. Lygus injury and tip burn can be readily distinguished if one becomes familiar with both.

DISTRIBUTION

Tip burn has been observed on sugar beets in the field in California, Oregon, Washington, Idaho, Utah, Colorado, and Arizona. It probably occurs wherever sugar beets are grown, the extent or amount varying with local conditions. On mother beets in the greenhouse it has been seen or reported in Utah, Wyoming, Colorado, Minnesota, and California.

IMPORTANCE

The economic importance of sugar-beet tip burn as a disease is almost negligible. It usually occurs as merely a temporary disturbance in large, vigorous beets. It has not been observed, as is sometimes the case with

lettuce tip burn, to open the way for secondary rotting organisms of importance. It does sometimes objectionably retard development of mother beets being grown in the greenhouse for breeding purposes.

EXPERIMENTAL WORK AND RESULTS

High Nitrogen Fertility

Many field observations by the junior author, beginning in 1917, on the occurrence of tip burn under conditions of high fertility led to the conclusion that the trouble is associated with very vigorous growth.[2] Repeated efforts were made in the field and the greenhouse to induce or in-

FIG. 2. Left. Tip burn of sugar beet experimentally induced by high nitrogen fertilization in the field followed by growing the plant in greenhouse under strongly reduced light. Several leaf blades are severely truncated. Large leaf at upper right shows puffy or swollen appearance of mesophyll tissue between larger veins. Photographed January 4, 1944. Right. Tip burn on end of sugar-beet seed stalk showing blackened tip of central stalk and several adjacent petioles with blackened tips but without blades. Normal young leaves on rudimentary axillary shoots are forerunners of recovery. Plant grown over winter in field at Corvallis, Oregon, and transplanted to cool greenhouse in Salt Lake City, Utah, on March 10, 1944. Photographed April 15, 1944.

[2] Charles Price, of this same field station, observed in 1932 at Hemet, California, that tip burn was more prevalent and more severe in experimental plots that received heavy applications of nitrogenous fertilizer than in a nearby commercial field in which little or no commercial fertilizer was applied. He also observed at Chino, California, the same year, that tip burn was severe on nearly all beet plants growing in an abandoned feed yard; whereas, only a small percentage of the beets growing immediately outside this area were affected and the symptoms were relatively mild.

crease the severity of tip burn by heavy nitrogen fertilization in various ways. These gave negative or inconclusive results, though in some instances symptoms thought to be precursors of definitely diagnostic symptoms were noted. The negative results of various other types of nutritional experiments together with the fact that complete recovery consistently occurs without artificial treatment indicated that tip burn is not due to a nutrient deficiency but rather to normal constituents of the plant temporarily present in such high concentrations as to be toxic.

FIG. 3. Recovery from tip burn. Tip and terminal leaves of apical flowering shoot checked in growth and severely injured. Two axillary shoots at sides are developing normally. Plant overwintered in field at Corvallis, Oregon. Transplanted to cool greenhouse at Salt Lake City, Utah, March 10, 1944. Photographed April 15, 1944.

The relation of nitrogen nutrition or high soil fertility to tip burn was tested at Riverside, California, in an experiment started October 10, 1941. Thirty-six plants of the self-fertile, uniform beet variety, S.L. 68, had been obtained in fine condition. These came from the curly-top-resistance breeding field, conducted by Albert M. Murphy of this same Division, near Buhl, Idaho. The plants were from a spring planting on soil of medium fertility.

Ten of these plants were planted in ordinary potting soil and placed in

a warm greenhouse. Thirteen of the plants were set out in an outdoor bed in unmanured soil of low fertility. The other thirteen were planted in another outdoor bed with similar exposure but in fertile soil heavily manured.

On November 13, thirty-four days after the beets were set out, the plants in the greenhouse were developing normally. Growth of the plants in the two outdoor beds had been retarded by cool weather. Some of the plants in the unmanured bed showed "blackening of leaf edges suggestive of tip burn." The plants in the manured bed had more blackened tissue along the leaf edges and were slightly darker green. Apparently symptoms of tip burn developed in both outdoor lots but to a somewhat greater degree in the highly fertile soil.

On March 13, 1942, two of the thirteen plants in the low-fertility bed had tip-burn symptoms. Five of the thirteen plants in the high-fertility bed had tip-burn symptoms.

On April 19, none of the plants in the low-fertility bed had tip-burn symptoms. Two of the plants in the high-fertility bed had definite tip-burn symptoms.

The evidence indicates that when the plants were set out some of them contained some substances in adequate amounts to give rise to tip burn. The evidence also indicates further accumulation of the tip-burn-inducing substances, when the plants were grown in soil of high fertility. Recovery from tip burn was faster in the low-fertility soil.

A somewhat comparable test with two levels of soil fertility in the same two outdoor beds was conducted with the variety S.L. 68 in 1943–1944. Seed was planted in the two beds on October 30, 1943. On January 26, ten plants from each bed were transplanted to 8–inch pots and then on February 18 these potted plants were placed in the greenhouse. Some of these plants from both fertility levels were given an extra treatment with ammonium sulphate.

On March 13, 1944, slight but definite tip-burn symptoms were noted on one plant in the high-fertility bed and on March 20 two more of these plants had slight, definite symptoms. None of the plants in the low-fertility bed and none of those in the greenhouse were so affected.

This test also affords support for the idea that, with other environmental influences equal, soil fertility or nitrogen nutrition is a factor in the development of tip burn.

Light Intensity

Relatively low light intensity[3] as a factor in tip burn development in heavily fertilized beets was demonstrated by the senior author in the spring

[3] After the discovery of the light relationship to tip burn was made it was learned through personal communication with E. L. LeClerg, formerly Assistant Pathologist of the Division of Sugar Plant Investigations, that while stationed at the University of Minnesota in 1937 he had experimentally induced tip burn in mother beets. He accomplished this by growing the plants in a chamber with high humidity and low light intensity.

of 1943. That low light intensity is a faotor was suggested by the fact that many mother beets and stecklings planted for another purpose at Riverside, California, on February 3, 1943, had developed tip burn following prolonged cloudy weather. After the weather cleared no more tip burn developed. Another suggestive consideration was the fact that the disease had been observed in this locality more commonly in the spring, when there is a good deal of cloudiness, than in the clearer weather of summer.

The low-light-intensity hypothesis was tested in a field experiment started May 19, 1943. The beets used were in a short row of the variety U.S. 15 in a plot planted November 7, 1942, for another purpose by Charles Price of this Division. The plants had been given uniform cultural care, up to April 22, 1943, and had been fertilized with sodium nitrate at about 250 pounds per acre on January 11 and again on March 18. On April 22, a heavy application of ammonium sulphate was made to one third of the row; a similar amout of calcium nitrate was applied to a second third of the row; the remaining third of the row received no additional fertilizer. The fertilizer applications were repeated on April 26.

Four plants were defoliated in the section of row normally fertilized and four in each of the two sections heavily fertilized. Two defoliated beets of each group of four were left in full sunlight. The other defoliated plants of each group were subjected to greatly reduced light intensity by inversion of a five-gallon paper ice-cream carton over each plant. Two openings 3×5 inches had been cut in the sides of each carton near the bottom. These opening permitted some ventilation but were coverd with dark cloth to exclude most of the light.

Tip-burn symptoms began to appear on the shaded, defoliated plants after ten days. Finally, all four of the shaded, heavily fertilized plants developed tip burn. Those that had received ammonium sulphate were more severely affected than those that had received calcium nitrate. The two shaded beets that received only normal fertilization did not develop tip burn. No tip burn occurred in the exposed, defoliated plants in all three sections of the row, nor in the nondefoliated plants in this row and in other parts of the field.

Other beets from the same experimental row in the field were used in a second light-relationship experiment in the greenhouse. The plants were removed from the field on May 19, held under uniform conditions of cold storage until June 5, and then planted in pots in the greenhouse. Some of the plants were exposed to full sunlight while others were held in dense shade as in the field experiment. The plants that had been taken from the section of the row receiving only normal fertilization did not develop tip burn whether exposed or shaded. The plants from the two sections given additional fertilization did not develop tip burn if exposed to the full sunlight but did develop the disease if shaded.

The results of these two experiments prove that under conditions involved low light intensity was a controlling factor in the causation of tip

burn. The results also support and confirm the earlier evidence that nitrogen nutrition is a factor in tip-burn causation.

Additional field and greenhouse experiments were conducted to confirm the evidence that low light intensity is a factor in the development of tip burn. The results with those of the first two experiments, proved that with beets preconditioned by growing under conditions of high nitrogen fertility tip burn will develop if the beets are then grown under very low light intensity. The results of these experiments are summarized in table 1.

Plants fertilized with ammonium sulphate and exposed to full sunlight for six days and then placed in reduced light failed to develop tip burn. Those similarly fertilized and exposed to full sunlight for three weeks and then placed in reduced light developed the disease.

When plants were shaded for two months after severe symptoms of tip burn had developed, the symptoms on the leaves appearing later were pro-

TABLE 1.—*Influence of low light intensity and nitrogen nutrition on sugar-beet tip burn*

Nitrogen nutrition preconditioning	Place of tests	Light intensity			
		Full sunlight		Reduced sunlight	
		Plants treated	Plants diseased	Plants treated	Plants diseased
		No.	*No.*	*No.*	*No.*
Moderate	Greenhouse	8	0	8	0
	Field	8	0	8	0
High	Greenhouse	59	0	59	57
	Field	7	0	7	7

gressively milder until finally leaves developed showing no evidence of tip burn. This suggests that the substances involved in the development of tip burn may be present in the plant in limited amounts. It also indicates that when these substances are depleted the plants again develop normally, even though they are still in a reduced light intensity.

Reduction of the light intensity was effected in other ways in addition to the use of the paper ice-cream cartons. Unbleached muslin bags, cheesecloth cages, and well-ventilated cardboard cartons were used. The light intensity under the unbleached muslin bags, sufficiently low to induce severe tip burn, was only 15 per cent of the full sunlight intensity on a clear day. This was determined with a pyrheliometer at the weather station of the University of California Citrus Experiment Station. The light target of the pyrheliometer was covered with an unbleached muslin bag for five minutes during a period of maximum light intensity on a clear day. Equilibrium was established within three minutes. The readings taken immediately before and after the instrument was shaded were the same. The ice-cream cartons, according to measurements with the same instrument, seemed to eliminate nearly all light, but the fact that they admitted a little light is evident because some chlorophyll developed in leaves of plants

covered by the cartons. Pyrheliometer records made by the same instrument on heavily overcast days in February and March showed that the light intensity for fully exposed plants was slightly lower for short periods of some days than under the muslin bags on bright days.

Genetic Variability

Genetic variability also appears to be a factor in the response of sugar beets to the environmental influences conducive to tip burn. It has been consistently observed in fields with uniform fertilization and general cultural care that individual plants with tip burn are distributed at random. All varieties of sugar beets now in commercial use are highly heterozygous. Further evidence of the relation of genetic composition to tip burn has been afforded by the fact that among breeding stocks and commercial varieties studied under uniform environmental conditions there will be more cases of tip burn among the more vigorous or yield-type varieties, such as U.S. 14, than among the less vigorous or sugar-type varieties, such as U.S. 15.

Alternating Light Intensities

The relation of alternation of light intensities to the development of tip burn and to recovery from the disease was tested experimentally. Plants that had been predisposed to tip burn from the standpoint of nitrogen nutrition were used. Such plants were alternately held for periods of two weeks or more in full sunlight and reduced light. (Table 2). Beets that

TABLE 2.—*The relation of alternating high and low light intensities to development of and recovery from tip burn*

Nitrogen fertilization treatment	No of plants treated	No. of plants with tip burn after successive treatments[a] at different light intensities					
		Full light	Reduced	Full light	Reduced	Full light	
Moderate	8	0	0	0	...	...	
Heavy	11	0	11	0	...	...	
Heavy	5	0	5	0	5	0	

[a] The plants were held continuously under a given light intensity for 14 days or longer.

had accumulated nitrogen reserves developed tip burn in low light intensity then recovered in high light intensity and then again developed tip burn if again held under low light intensity.

Food Reserves

The substances which are involved in the toxic reaction in tip burn probably accumulate with other reserves in the beet root. These substances manifest themselves through tip burn on large well-nourished beets when such plants are defoliated and shaded enough to retard or preclude photosynthesis. Under such circumstances the toxic substances would appear to come from the roots. The fact that some defoliated beets that were grown in sphagnum moss were noted by Bennett of this Division to show tip burn

on the new leaves supports the idea that the toxic substances arise directly or indirectly from storage products in the roots.

Movement of the curly-top virus in beets has been shown by Bennett (5) to be in the phloem and in the direction of flow of elaborated food. If the movement of food in a beet in reduced light is the result of mass flow of the fluid contents of the phloem from a point of high pressure (the root) to one of lower pressure (the top) any toxic substances present would move up with the sugars.

Experiments were conducted to test the hypothesis that tip burn is dependent on the direction of flow of food in the plant. Six large beet roots were potted and heavily fertilized. They were grown in full sunlight in the greenhouse until 15 or more leaves had developed. The three smallest inner leaves were then shaded with a muslin bag. The bag was so adjusted that all new leaves coming out later would be inside it. The remainder of the leaves were left on the plant and exposed to full sunlight. When 12 to 16 leaves, all healthy, had developed in the reduced light under the muslin bags, adequate time for tip burn to develop, all the outer leaves and all but six of the smallest leaves which had developed under the bag were removed from the test plants. The six smallest leaves were kept in reduced light. The plants were kept shaded until 10 to 14 additional leaves had developed under the reduced light. All plants developed tip burn. The shades were then removed and the plants exposed to full sunlight. All plants then recovered. A control plant of the same lot left continuously in full sunlight remained healthy.

A second test, in which 9 plants were used, was conducted in a similar manner. Cloudy weather prevailed for two weeks after the roots were potted and as a result all the plants had mild symptoms of tip burn on the first leaves that came out. This showed that all the plants were capable of developing tip burn. The weather then cleared and 16 to 20 new healthy leaves developed in full sunlight. The three smallest leaves of each plant were then shaded with a muslin bag. The plants were kept shaded until 10 to 14 additional leaves had developed under reduced light. The outer leaves were removed from 5 plants while the remaining 4 plants served as controls. Tip burn developed on the plants from which the outer leaves had been removed. No tip burn developed on the controls. Then the leaves of the 4 control plants were also removed. These all then developed tip burn. Thus, a total of 9 plants developed tip burn when the center leaves were shaded and the outer leaves removed.

Two experiments were conducted to demonstrate that a sufficient reduction in the light intensity for tip burn to develop was also sufficient to force the plant to draw on the reserves stored in the root. In the first test 20 beets ranging in weight from 216 to 475 grams were dug from a field plot and defoliated. The sucrose, determined on cores removed from the roots, averaged 14.8 per cent. The roots were then planted in 8-inch pots and shaded sufficiently to induce severe tip burn. The beets were left shaded

until about 20 leaves had appeared on each beet. The shades were removed and a second core removed from each beet for sucrose determinations. Sucrose at the end of the test averaged 8.4 per cent. During the period the beets were shaded they lost an average of 43 per cent of their total sucrose. All but 6 of the 20 beets had severe symptoms of tip burn. Of the 10 largest beets (288 to 475 grams) 8 developed tip burns, whereas only 6 of the 10 beets smaller than 288 grams developed symptoms of tip burn. This evidence suggests that there may be a correlation between beet size and development of tip burn.

This experiment was repeated in the field, shading approximately 100 beets in all. Every other beet in a section of 4 rows was defoliated and a core removed from the roots with a cork borer. The beets were then shaded with a fine weave cheesecloth. This cloth when clean reduced the light intensity 68 per cent when placed over a pyrheliometer on a clear day. After 12 to 16 leaves had developed on the defoliated plants the shades were removed and cores removed from both the defoliated roots which were sampled at the beginning and also from the undefoliated roots.

In this test the reduction in the amount of light by shading was sufficient to cause only a 10 per cent loss in sucrose from the roots of the undefoliated plants and a 26 per cent loss in the sugar content of the defoliated plants. Mild though distinct symptoms of tip burn developed on approximately 25 per cent of the shaded plants. Mild cupping of the leaves, an incipient stage of tip burn, was observed on approximately 25 per cent more of the shaded plants. The weights of these roots were not known, but it is certain that many roots were small. The size relationship in conjunction with the higher light intensity used may explain the low percentage of the plants which developed tip burn. No tip burn was observed in the remainder of the plot during the experiment.

DISCUSSION

The fact that tip burn develops in the field during prolonged periods of cloudy or foggy weather suggests the need to consider humidity as a possible factor. The possibility of a temperature relationship should also be considered. Relative humidity, at least as it prevailed in the greenhouse, appeared not to influence the development of tip burn in the test plants used. In view of the wide range of temperature under which tip burn has been observed to develop, there appears to be no critical range of temperature required.

The substances that cause sugar-beet tip burn are probably nitrogenous compounds but since the injury does not appear immediately after plants are heavily fertilized and shaded the effect is likely not due to the ammonium or the nitrate ion. The injury is probably due to complex nitrogenous compounds intermediate between the ammonium ion and the proteins.

If, as is apparently the case, the toxic substances accumulate in the roots and if they move up with the food in the phloem, a hypothetical ex-

planation can be advanced as to why tip burn did not develop on plants with the younger, inner leaves in reduced light and the outer leaves in full sunlight. In such cases perhaps the exposed outer leaves would supply the food requirements of the covered leaves so that there would be little or no movement of substances from the root to the covered leaves through the phloem.

Another hypothetical explanation of the fact tip burn did not develop on the covered inner leaves when the outer leaves were exposed to full sunlight is that perhaps products of photosynthesis combine with the toxic substances and in effect neutralize them. Under such conditions perhaps the substances that cause tip burn would be reduced to nontoxic concentrations.

If the tip burn is due to translocation of substances in the phloem from the root to the tops under reduced light then it would follow that recovery from tip burn under full sunlight would be due to a reversal of the direction of the flow of the phloem contents. If, on the other hand, tip burn is due to substances not adequately neutralized by products of photosynthesis under reduced light, then one could postulate that the increased amount of photosynthates under high light intensity would hold the toxic substances below the critical concentration.

SUMMARY

Tip burn of sugar beet is a disease believed due to toxic concentrations of substances normally present in the plant.

Tip burn develops when beets that have been grown in fertile soil with an abundance of nitrogen for a relatively long time are then grown under a low light intensity. The nitrogen and the light factors are both necessary.

When the outer leaves are left on the beet plant and in full sunlight tip burn does not develop on the new leaves even though they are shaded.

The toxic substances involved in tip burn are apparently accumulated in the root and are probably normal nitrogenous constituents of the plant temporarily in excessive concentrations.

Complete recovery from tip burn occurs if affected beets are grown under high light intensity and conditions otherwise favorable for beet growth.

Sugar beets are, in general, highly heterozygous and vary widely in their sensitiveness to the complex of factors causing tip burn.

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LITERATURE CITED

1. BONCQUET, P. A. *Bacillus morulans* n. sp. a bacterial organism found associated with curly top of the sugar beet. Phytopath. **7**: 270–289. 1917.
2. ROBBINS, W. W. Mosaic disease of sugar beets. Phytopath. **11**: 349–365. 1921.
3. LUTMAN, B. F. Tip burn of potato and other plants. Vermont Agric. Exp. Sta. Bull. 214. 1919.
4. BALL, E. D. The potato leafhopper and the hopperburn. Phytopath. **9**: 291–293. 1919.
5. BENNETT, C. W. Correlation between movement of the curly top virus and translocation of food in tobacco and sugar beet. Jour. Agr. Res. [U. S.] **54**: 479–502. 1937.

PATHOGENICITY OF THE VASCULAR FUSARIUM OF GLADIOLUS TO SOME ADDITIONAL IRIDACEOUS PLANTS

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Members of the Iris family are frequently grown in association by both commercial and amateur growers. With *Fusarium* rapidly increasing in importance in Gladiolus production the question arose as to whether or not plants of related genera in the Iridaceae would become infected with the vascular *Fusarium* from *Gladiolus* recently described by McCulloch (4) as *Fusarium orthoceras* App. and Wr. var. *gladioli*. Since 1941 plants of 11 genera in the Iridaceae have been inoculated with isolates Nos. 12–1, 16–1, and 19–1 from Miss McCulloch's collection, with positive infection resulting in plants of 9 genera and possible infection in plants of 2 genera.

MATERIALS AND METHODS

The plant materials tested were *Homeria collina* Vent., *Neomarica gracilis* Herb., *Streptanthera cuprea* Sweet, *Tritonia crocata* Ker., *T. lineata* (Salisb.) Ker., *Babiana* hybrids, *Crocus* (spring-flowering, mixed), *Ixia* hybrids ("Bloem Erf," "Dutch," and "Mrs. Cleveland's"), *Sparaxis* (assorted), and *Watsonia* (assorted), all from commercial sources. Bulbous iris varieties Imperator, Poggenbeek, and Wedgewood were from stock grown at the Plant Industry Station, Beltsville, Md., and corms of *Freesia* were from seedlings grown in sterilized soil at the Station. Corms of *Tritonia crocata* used in 1944 were also seedlings grown at Beltsville.

In the 1941–42 tests all of the bulbs and corms were grown in sterilized soil in 5-inch pots and, with the exception of the bulbous *Iris* and the *Watsonia*, were planted 3 to a pot. In the 1942–43 and 1944 tests sterilized sand was used and the plants were fed weekly with a nutrient solution. In these tests the plants, with the exception of bulbous *Iris*, were grown in 4-inch pots. The bulbous *Iris* was grown in flats of sterilized sand. The *Iris* was planted 24 per flat, the *Watsonia* and *Crocus* 1 per pot, and other genera 3 per pot.

Some of the corms and bulbs were infected with *Fusarium* when they were planted, but the likelihood of contamination was kept at a minimum by the placing of all pots in sterilized saucers, by a wide spacing of the pots, and by care in watering. In spite of these precautions some contamination occurred, chiefly in 1941–42 when the space used was greatly reduced to provide room for war emergency crops. While the plants used in these experiments were being moved into smaller space the pots were allowed to come in contact with each other.

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The 1941–42 inoculum was prepared from isolates 12–1, 16–1, and 19–1 grown on sterile oats for 2 weeks. Inocula of the 3 isolates were mixed and about 40 cc. of the mixture placed over some soil in the bottom of each pot. The inoculum was covered with soil, the bulbs or corms were then planted, and the pot was filled with soil. An equal amount of sterile oats was placed in each uninoculated pot.

TABLE 1.—*Infection of representatives of 11 genera of the Iridaceae with the vascular Fusarium from Gladiolus*

Plants tested	Year	Uninoculated: No. of bulbs, corms, rhizomes	Uninoculated: Fusarium isolated[a]	Inoculated: No. of bulbs, corms, rhizomes	Inoculated: Fusarium isolated[a]	Type of infection in bulbs, corms, rhizomes
Babiana, hybrids	1941–42	3	0/1	9	4/5	Vascular
	1942–43	6	0/6	18	3/18	do
Crocus (mixed, spring-flowering)	1944	6	0/6	18	13/18	do and basal
Freesia, seedlings	1941–42	15	1/3	45	7/23	Vascular
	1942–43	18	1/17	40	33/43	do
	1944	9	0/9	15	3/15	do
Homeria collina Vent.	1941–42	3	0/1	9	1/5	do and basal
Iris (bulbous) var. Imperator	1941–42	4	1/3	14	1/12	Basal
	1942–43	24	1/19	48	9/34	do
var. Poggenbeek	1942–43	24	3/20	48	13/21	do
var. Wedgewood	1941–42	4	0/4	12	7/9	do
	1942–43	24	7/23	48	18/29	do
Ixia, hybrids, "Bloem Erf"	1941–42	3	2/2	9	6/7	Vascular
	1942–43	3	0/3	9	2/9	do
"Dutch"	1941–42	3	1/1	9	5/6	do
	1942–43	3	0/3	9	8/9	do
"Mrs. Cleveland's"	1941–42	3	1/2	9	6/6	do
	1942–43	3	0/3	9	6/9	do
Neomarica gracilis Herb.	1941–42	1	0/1	2	1/2	No symptoms
Sparaxis, assorted	1941–42	3	0/2	9	6/9	Vascular
	1942–43	6	0/6	18	12/18	Basal
Streptanthera cuprea Sweet	1941–42	3	?	9	5/7	Slight vascular in base
	1942–43	3	0/2	9	9/9	do
Tritonia crocata Ker., seedlings	1941–42	4	0/2	18	1/3	Vascular
	1944	9	0/9	15	3/15	do
T. lineata (Salisb.) Ker.	1942–43	3	0/3	9	0/6	No symptoms
Watsonia, assorted	1941–42	4	0/4	16	6/16	Vascular and basal
	1942–43	10	0/5	29	4/9	do

[a] The numerator indicates the number of successful isolations; the denominator, the number of bulbs from which isolations were made.

For the 1942–43 and 1944 tests each of the three isolates was grown in a modified Richard's solution and ground in a Waring Blendor. The bulbs and corms were dipped just before planting in a mixture of the three isolates.

Isolations were made from many of the corms or bulbs when recognizable foliage symptoms appeared. In many instances the corms or bulbs had

rotted so badly that isolations were not attempted. Isolations were made from most of the remaining bulbs at maturity.

Representative isolates recovered from inoculated plants of each susceptible genus except *Crocus, Homeria, Neomarica,* and *Tritonia* have been tested for pathogenicity in *Gladiolus.* Twelve corms each of the *Gladiolus* varieties Picardy and Dr. F. E. Bennett were dipped in a suspension of a mixture of representative isolates from a given genus and planted in sterilized sand in 8-inch pots, 3 corms to a pot. These corms were fed weekly with a nutrient solution.

RESULTS

The results of the inoculations are in table 1. Since isolation was usually attempted from the vascular system or the base of the corm or bulb but seldom from the roots, *Fusarium* was not always recovered from plants which were obviously diseased. Positive infection was obtained in the following: *Babiana, Crocus, Freesia,* bulbous *Iris, Ixia, Sparaxis, Streptanthera, Tritonia,* and *Watsonia.* Typical reddish-brown vascular discoloration occurred in one or more bulbs or corms in each of these. In *Neomarica,* however, no recognizable symptoms were present in roots, tops, or rhizomes. Whether true infection existed in *Neomarica* is open to question. Rotting of the roots and corm or bulb base was a general symptom of the effects of this *Fusarium* in all genera. This effect was the predominating symptom complex in the bulbous *Iris* whereas the characteristic symptom in *Crocus, Ixia,* and *Watsonia* was the typical reddish-brown vascular discoloration. Detailed symptoms are described.

Babiana.—The thin leaves became yellow and died back from the tips leaving the dead, yellow-brown tissue thin and paper-like. This necrosis gradually proceeded down the leaf towards the corm. Plants remained alive in the greenhouse as long as 5 months after inoculation. The central vascular cylinder was discolored if infection had progressed sufficiently. Frequently this brown discoloration was confined to the base of the vascular cylinder but in one plant it extended throughout the central cylinder and into the stem. In this plant the parent corm appeared to be normal, the fungus entering the daughter corm through the contractile roots.

Crocus.—Foliage symptoms first appeared within 4 weeks after inoculation on one of the plants and were present within 9 weeks in all but one of those inoculated. The first symptom, a slight yellowing at the tips of the leaves, was soon followed by necrosis as the yellowing progressed down the leaves. Many of the leaves were twisted or curled (Fig. 1) and in 1 plant this curling was extremely severe. Stunting also occurred. One plant grew very little and was nearly dead 5 weeks after inoculation. The roots usually rotted off; thus the absence of roots (Fig. 1) and the presence of basal corm rot were distinguishing symptoms. This light brown basal rot sometimes involved the lower half of the corm, and brown node lesions were frequent in the corms. Vascular discoloration was absent

in most of the corms, but in 2 of these the discoloration extended through the parent corm into the daughter corm (Fig. 1).

The symptoms produced in *Crocus* by the Gladiolus *Fusarium* were very similar to those illustrated in *Crocus* by Drayton (3) and by Moore (5). It is probable that these workers were dealing with the same *Fusarium* as the writer. Drayton reported that Crocus rot was of serious consequence in Holland and that it is primarily a basal decay which advances further during storage and may completely rot the corm. Moore said that the Fusarium rot was the most prevalent and destructive disease of Crocus corms; and Pape (6) mentioned a Fusarium corm rot which frequently

FIG. 1. Crocus plants grown for 60 days in sterilized sand, fed weekly with a nutrient solution. The two plants at the left were not inoculated. Those at the right were inoculated by dipping the corms in a spore and mycelium suspension of the Gladiolus *Fusarium*.

completely destroyed the corms in storage. Attempts at infection of *Gladiolus* with the *Fusarium* obtained from *Crocus* were not reported by these writers nor was the possible relationship between the Gladiolus *Fusarium* and the Crocus *Fusarium* suggested. Abe (1) described a root and corm rot of *Crocus sativus* L. which is said to be caused by *Fusarium bulbigenum* Cke. et Mass. var. *blasticola* (Rostr.) Wr. According to the system of Snyder and Hansen (8) this would become *F. oxysporum* Schl. Abe was unable to infect onion seedlings with this *Fusarium*.

Freesia.—Freesias used in these tests have been very susceptible to the Gladiolus *Fusarium*. Yellowing of the foliage was evident within 30 days

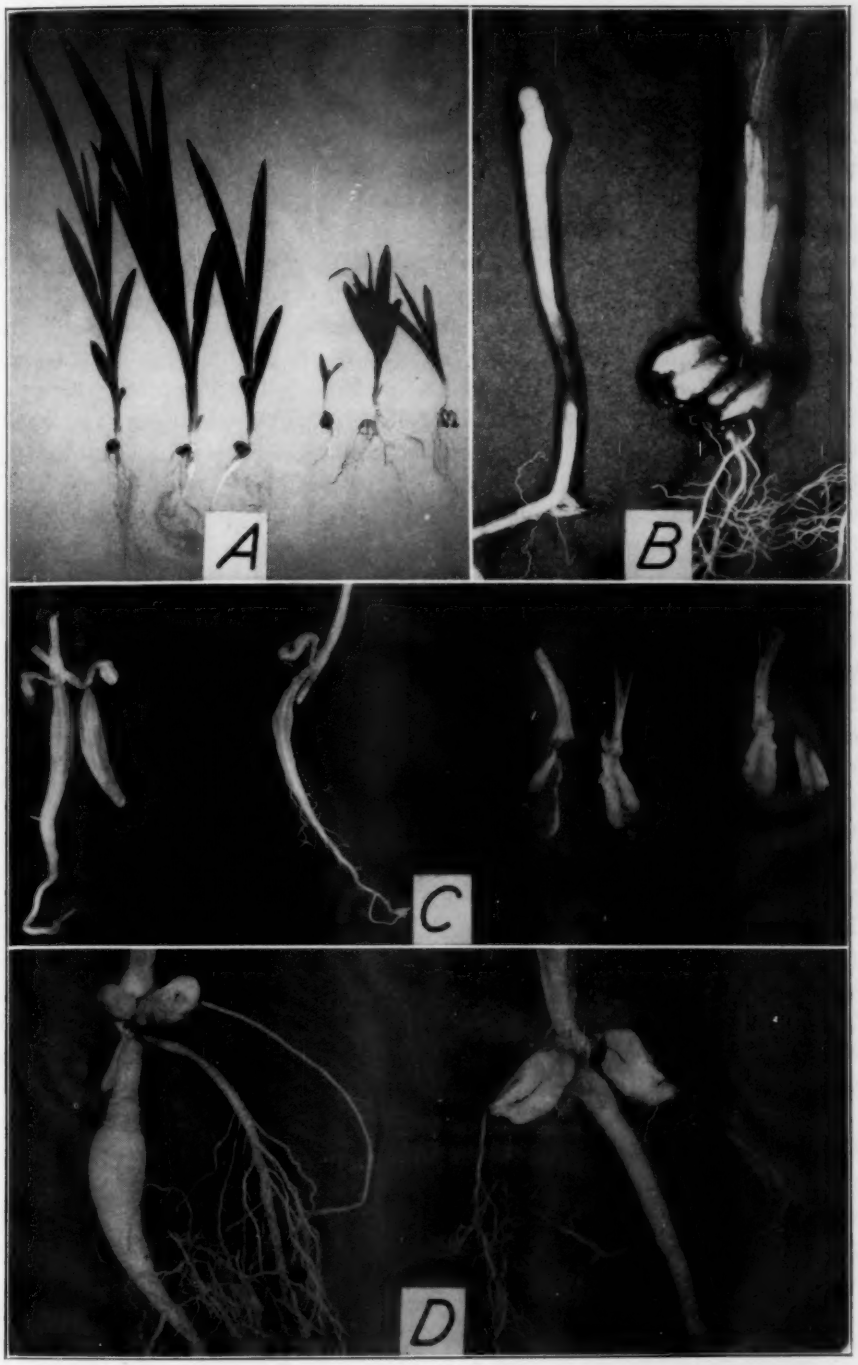

FIG. 2. Freesia plants grown in sterile sand, fed weekly with a nutrient solution. A. Right, plants 63 days after corms were inoculated by dipping them in a spore and mycelium suspension; left, not inoculated. B. Lesion on the main root (left); core rot of parent corm (right). C. Sections through healthy corms (left); diseased corms (right) showing basal and core rot. Plants are of the same age. D. Plants of the same age showing retardation of corm development following inoculation (right), compared with normal development (left). Note that the parent corm of the healthy plant has almost completely shriveled and that the large fleshy root is swollen.

after inoculation, and within 60 days many of the tops were completely dead and the corms rotted. The most pronounced effect of this *Fusarium* on *Freesia* was the stunting and retarding of the development of the entire plant. Even though no foliage yellowing was evident, growth was but one-third to one-half that of uninoculated plants (Fig. 2, A). The shriveling of the parent corm and the swelling of the main root was greatly retarded in inoculated plants (Fig. 2, D). The first symptom, in addition to stunting, was a yellowing and dying back of the foliage. Frequently the leaf tips were white rather than yellow, similar to a physiological burn. This die-back was accompanied by a decay of the root tips, by root lesions (Fig. 2, B), by vascular discoloration, and finally by complete rotting of the corms (Fig. 2, B and C). The infected central vascular core was either brilliant red or dark brown, this discoloration frequently extending through both the parent and daughter corms into the leaf bases. Occasionally, also, the radiating vascular strands were discolored. As the rot progressed the base of the corm rotted and surface lesions appeared elsewhere on the corm.

Taubenhaus and Ezekiel (9), reporting a Fusarium wilt and corm rot of *Freesia,* described and figured symptoms similar to the above but they did not mention a retardation in the development of the daughter corm. They reported only slight decay of the basal plate and very little stunting when Freesia corms were inoculated with a *Fusarium* from decayed Gladiolus corms, whereas isolates of *Fusarium* from *Freesia,* tomato, and cabbage were reported as causing a typical core rot. Corms of *Gladiolus* that were planted in soil infested with the Freesia *Fusarium* showed a slight decay at the bases which prevented the formation of new roots. As McCulloch (4) has pointed out, their failure to obtain infection may have been due to the use of varieties which were resistant to the strain of *Fusarium* used in the tests. Similarly McCulloch (4) was unsuccessful with cross inoculations with Fusaria from *Gladiolus* and from *Freesia,* and she believed her failure may have been due to the use of resistant varieties as test plants. She stated that the Fusarium-infected Freesia corms had symptoms similar to the vascular disease of *Gladiolus* and that the *Fusarium* isolated from these corms appeared to be identical with her Gladiolus *Fusarium.*

Homeria collina.—The symptom complex has not been well established for this species. As observed, the leaves died back from the tips and, when plants were lifted for examination, the corms looked healthy but all the basal roots were either dying back or had rotted away. Many of the contractile roots were brown at the tips. Only slight vascular discoloration was seen in the bases of 2 corms nearly a year after inoculation.

Iris (bulbous).—One or more species of *Penicillium* appear to be closely associated with Fusarium infection in bulbous Iris. Many Iris bulbs first inoculated and infected with *Fusarium* soon had *Penicillium* fruiting freely between the rotting bulb scales. No doubt Fusarium rot of bulbous Iris is frequently confused with the Penicillium rot described by Moore (5). Fusarium infection was characterized by a stunting of the plant and by the darkened bulb bases and the absence of roots (Fig. 3). If secondary in-

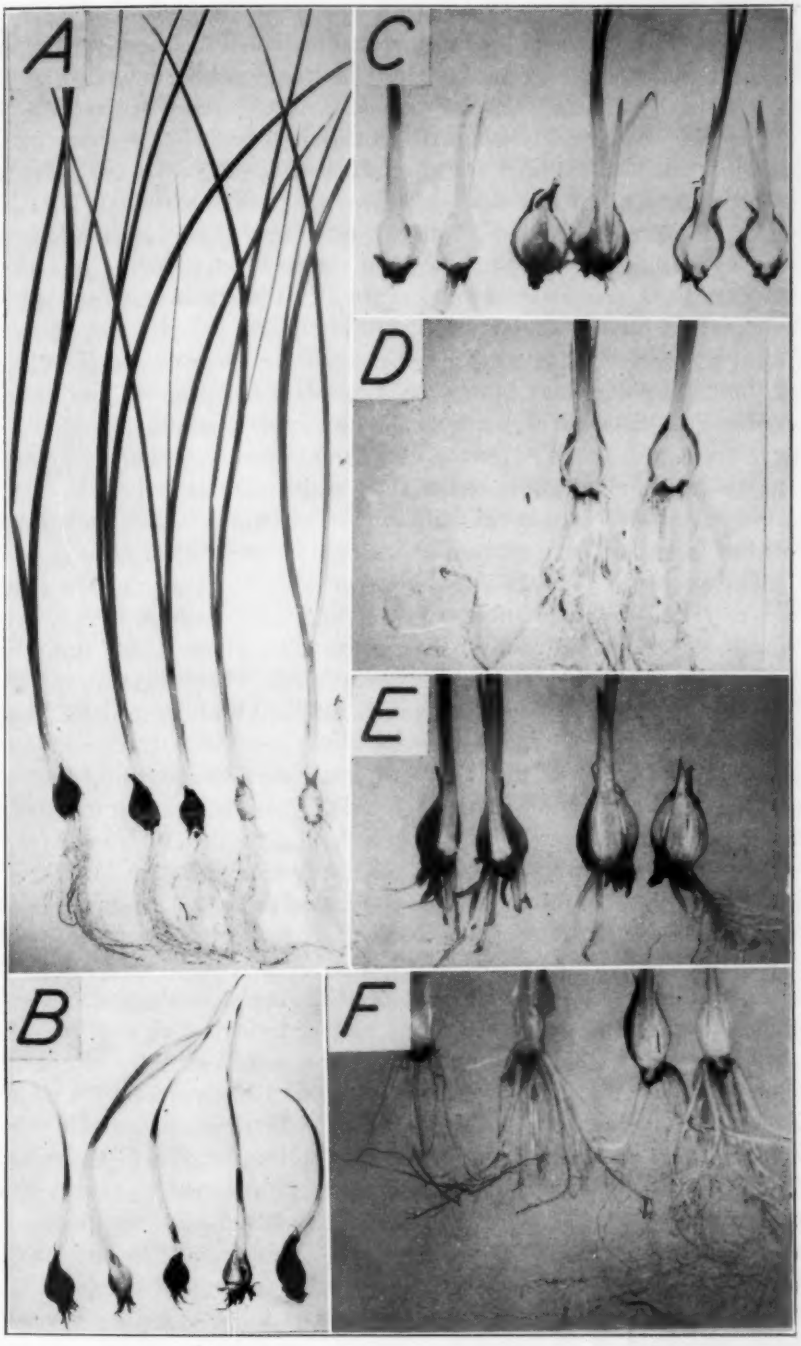

FIG. 3. Effect of the Gladiolus *Fusarium* on bulbous Iris. All plants are the same age. A–B. Wedgewood not inoculated (A), inoculated (B). Note the difference in growth and the absence of roots. C–F. Longitudinal sections through diseased and healthy Imperator (C and D), and Poggenbeek (E and F), showing the discolored bases and bulb scales in the inoculated bulbs (C and E).

fection by *Penicillium* did not occur the bulb remained firm with a badly discolored and rotted base. Browned vascular tissues frequently were noted in the bulb scales and occasionally vascular discoloration extended into the stem, but usually the discoloration was confined to the base of the bulb (Fig. 3). *Fusarium* was readily isolated from such areas. Roots were few and poorly developed or had been rotted off altogether (Fig. 3). No differences in susceptibility of the 3 varieties tested were noted although F. P. McWhorter, in a personal communication, stated that the Poggenbeek variety is severely affected by a Fusarium basal rot in the field, and Creager (2) stated that a *Fusarium* is very destructive to the Cajanus, Hart Nibbrig, and Imperator varieties. A mixture of bulbs of Wedgewood and other varieties was sent the writer from Grants Pass, Oregon, by McWhorter. These bulbs had darkened bulb bases typical of those inoculated with the Gladiolus *Fusarium*. A *Fusarium* similar to the Gladiolus *Fusarium* was isolated from both the Wedgewood and David Haring varieties of *Iris*, but no infection was obtained in either the Picardy or the Dr. F. E. Bennett variety of *Gladiolus* with either isolate. A *Fusarium* isolated from bulbous *Iris* from Texas likewise produced no infection in these 2 varieties of *Gladiolus*. This discrepancy in behavior between the Gladiolus and the Iris isolates is as yet unexplained but may be due to differences in behavior between strains of *Fusarium*. Strains of the Gladiolus *Fusarium* differing in pathogenicity are known to exist (unpublished data).

Creager (2) has published a note on a root and bulb rot of both Spanish and Dutch *Iris* which is caused by a *Fusarium* "closely resembling *F. oxysporum*." He stated that the pathogen enters the root and finally involves the basal plate and scales of the bulb. No mention was made of any cross-inoculation studies.

Ixia.—The first symptom of Fusarium infection was a yellowing of the tips of the leaves. As this yellowing progressed down the leaves these began to twist and their tips died. The roots of such plants were usually badly decayed and the vascular system of the contractile roots sometimes was discolored. In later stages all or nearly all of the roots were rotted. The base of the corm was frequently rotted and surface lesions were present. Basal rotting gradually extended upward into the central vascular cylinder, thus producing a core rot. The radiating vascular tissues were likewise often discolored. As the disease progressed it sometimes became a general corm rot and often a corky dry rot followed the discoloration of the radiating vascular tissues. Brilliant red areas frequently defined the corm lesions, but apparently the red color is not always associated with the disease since it also occurs as a result of other injuries. The daughter corm usually became infected through the connecting vascular tissue, but occasionally the daughter corm became infected from contractile roots before the parent corm was invaded.

Neomarica gracilis.—No recognizable symptoms were observed, but *Fusarium* was obtained from one of two plants inoculated. It is questionable whether there was true infection.

Sparaxis.—Typical vascular and core rot was less common in *Sparaxis* than in *Freesia, Crocus,* or *Ixia.* The leaves yellowed and died back as in these other genera, and the roots were few, poorly developed and dying back, but often there was no vascular discoloration in either the fibrous or the contractile roots. Several corms, however, had definite vascular discoloration and core rot. The core of one parent corm was rotted and this decay extended slightly into the daughter corm. *Fusarium* was also isolated from a discolored area in the basal plate of another daughter corm.

Streptanthera.—The leaves became yellow and died at the tips, and the plants were badly stunted. The roots rotted off and the vascular core was discolored. The discoloration was yellow brown at first, but became much darker brown as the disease progressed. Some surface rot occurred at the base of the corm and eventually both parent and daughter corms rotted completely. *Fusarium* was isolated from such rotting tissue as well as from the lightly discolored and the darker vascular system.

Tritonia.—In the 1941–42 tests 12 of the 18 corms of *T. crocata* which were inoculated and planted rotted completely and none of them sent up new shoots, whereas all of the 4 uninoculated corms grew well and were in good condition when dug 11 months later. Of the 15 seedling corms of *T. crocata* planted in 1944, 3 had typical vascular discoloration and the others had none. *Fusarium* was isolated from these 3 corms. The foliage of all of the inoculated plants had begun to die back within 30 to 45 days after inoculation whereas the uninoculated plants retained a healthy appearance. Possibly distinct varietal resistance to *Fusarium* such as is known to occur in *Gladiolus* (4) also occurs in *Tritonia.* Plants of *Tritonia lineata* inoculated in 1942–43 remained healthy.

Watsonia.—The foliage of infected plants had a burned appearance around the edges and tips and then became yellow toward the base. Most of the roots rotted off although occasional healthy roots remained. The roots might or might not be entirely rotted away, with no evidence of corm rot other than slight rotting at the root bases. Usually, however, rotting of the roots was accompanied by a basal rot of the corm and occasionally the entire corm rotted. The vascular system of 1 corm was discolored throughout and extended into the stem.

REINFECTION OF GLADIOLUS

Successful reinfection of the very susceptible variety of *Gladiolus,* Dr. F. E. Bennett, was obtained with the Fusaria isolated from plants of the following genera: *Babiana, Freesia, Iris, Ixia, Sparaxis,* and *Watsonia.* Only slight vascular or basal infection of the Picardy variety was obtained with isolates from these genera. Slight vascular infection of Dr. F. E. Bennett, but not of Picardy, was obtained in 3 trials involving 42 corms of each variety with isolates from *Streptanthera.* Reinfection of *Gladiolus* has not yet been attempted with the isolates from *Crocus, Homeria, Neomarica,* or *Tritonia.*

DISCUSSION

The host range of the vascular *Fusarium* of *Gladiolus* is not confined to a single genus, but as yet these studies have not progressed far enough to determine whether Fusaria isolated from other members of the Iridaceae are pathogenic to *Gladiolus*. In limited trials isolates of *Fusarium* obtained from bulbous *Iris* were not pathogenic to *Gladiolus*. Recent investigations (7) have shown that some of the vascular Fusaria do not have the narrow host range attributed to them. This does not seem particularly surprising in view of the fact that Wellman and Blaisdell (10) were able to select some variants of *Fusarium bulbigenum* var. *lycopersici* (Brushi) Wr. and R. that were more pathogenic to tomatoes and some which were less. It would seem likely that it might be possible to select variants that would be pathogenic on closely related or even remotely related plants especially if we accept Snyder and Hansen's (8) concept of *F. oxysporum* Schl. and their statement that "the Fusaria which cause vascular wilts are merely biologic forms of one and the same species."

In view of the results obtained with the Gladiolus *Fusarium*, members of the Iridaceae should not be grown in association where the vascular *Fusarium* is known to be present.

SUMMARY

The vascular *Fusarium* of *Gladiolus* described by McCulloch proved to be pathogenic to plants of the following genera of the Iridaceae: *Babiana, Crocus, Freesia, Iris* (bulbous), *Ixia, Sparaxis, Streptanthera, Tritonia,* and *Watsonia*. Infection was uncertain in *Neomarica* and *Homeria*.

A *Fusarium* isolated from bulbous *Iris* was nonpathogenic to the Picardy and Dr. F. E. Bennett varieties of *Gladiolus*.

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LITERATURE CITED

1. ABE, T. Studies on a new dry-rot disease of the bulb of *Crocus sativus* L. caused by *Fusarium bulbigenum* Cke. et Mass. var. *blasticola* (Rostr.). Wr. Trans. Tottori Soc. Agr. Sci. **4**: 212–228. 1933.
2. CREAGER, D. B. Fusarium basal rot of bulbous iris. Phytopath. **23**: 7. 1933.
3. DRAYTON, F. L. Crocus diseases. *In* Report of the Dominion Botanist for the year 1928. Dominion of Canada Dept. of Agric., p. 22. 1930.
4. MCCULLOCH, LUCIA. A vascular disease of gladiolus caused by Fusarium. Phytopath. **34**: 263–287. 1944.
5. MOORE, W. C. Diseases of bulbs. Min. Agr. and Fisheries (Great Britain) Bull. 117. 1939.
6. PAPE, H. Crocus. Fusariumfäule der Knollen. *In* Krankheiten und Schädlinge der Zierpflanzen, p. 163. 1932.
7. SMITH, T. E., and K. J. SHAW. Pathogenicity studies with Fusaria isolated from tobacco, sweet potato and cotton. Phytopath. **33**: 469–483. 1943.
8. SNYDER, W. C., and H. N. HANSEN. The species concept in Fusarium. Amer. Jour. Bot. **27**: 64–67. 1940.
9. TAUBENHAUS, J. J., and W. N. EZEKIEL. Fusarium wilt and corm rot of freesias. Bot. Gaz. **95**: 128–142. 1933.
10. WELLMAN, FREDERICK L., and DOROTHY J. BLAISDELL. Pathogenic and cultural variation among single-spore isolates from strains of the tomato-wilt Fusarium. Phytopath. **31**: 103–120. 1941.

PHYTOPATHOLOGICAL NOTES

A Cross between Lycopersicon esculentum and Disease-Resistant L. peruvianum.—A group of varieties of *Lycopersicon peruvianum* (L.) Mill. and related species imported from Peru, South America, by the Division of Plant Exploration and Introduction, have had a most consistent resistance to tomato foliage diseases. Various workers report that lines have also been found with varying degrees of resistance to curly top, fusarium wilt, nematode rootknot, bacterial spot, and mosaic.[1] Because of this marked resistance of certain varieties of *L. peruvianum* to several tomato diseases, breeders have endeavored to hybridize them with varieties of *L. esculentum* Mill. in order to incorporate their resistance into new superior horticultural types.

Hybrids have already been reported between *Lycopersicon esculentum* and *L. chilense* Dun.[2] (*L. peruvianum* var. *dentatum* (Dun.)[3]), between *L. esculentum* and *L. peruvianum* var. *humifusum* C. H. Mull.,[4] and between *L. esculentum* and various species of the subgenus *Eriopersicon* C. H. Mull. group.[3, 5] However, so far as the writers have been able to ascertain, no cross between *L. esculentum* and the type species of *Eriopersicon, L. peruvianum,* has heretofore been reported.

When the work here recorded was started selections were made from introductions of *Lycopersicon peruvianum* grown in the field tests at Beltsville, Md. The plants that retained the healthiest foliage throughout the growing season were chosen as parent stocks. The pollen parent (Fig. 1, C, F, I) of the hybrid here reported is one of the more fruitful lines and bears large numbers of small greenish fruits usually tinted with purple when mature and averaging about $\frac{5}{8}$ inch in diameter.

After examining several hundred seedless tomato fruits that developed in the greenhouse at Beltsville, Md., from cross pollination of numerous tomato varieties with pollen of *Lycopersicon peruvianum,* viable seed was obtained in a cross with Prince Borghese, an Italian paste type. This variety of *L. esculentum* is characterized by the production of many small flowers on large forking cymes which bear large clusters of elongated, bright red, 2-celled fruits (Fig. 1, A, D, G).

The F_1 plants were vigorous vegetative growers and developed vines about twice the size of either parental stock. These plants flowered and produced pollen in the greenhouse but set no fruit unless a growth-regulating substance such as naphthalene acetamide or indolebutyric acid was

[1] Alexander, L. J., R. E. Lincoln, and V. Wright. A survey of the genus Lycopersicon for resistance to the important tomato diseases occurring in Ohio and Indiana. U. S. Dept. Agr., Plt. Dis. Reporter Suppl. 136. 1942.

[2] Holmes, F. O. The Chilean tomato, *Lycopersicon chilense,* as a possible source of disease resistance. Phytopath. 29: 215–216. 1939.

[3] Muller, C. H. A revision of the genus *Lycopersicon.* U. S. Dept. Agr. Miscellaneous Publication 382. 1940.

[4] Wright, V., and R. E. Lincoln. Resistance to defoliation disease in tomato. Purdue Univ., Agr. Exp. Stat., Ann. Rep. 53 (1940): 42–43. 1940.

[5] Blood, H. L. Curly top, the most serious menace to tomato production in Utah. Utah Agr. Exp. Stat. Farm and Home Science 3 (1): 8–11. 1942.

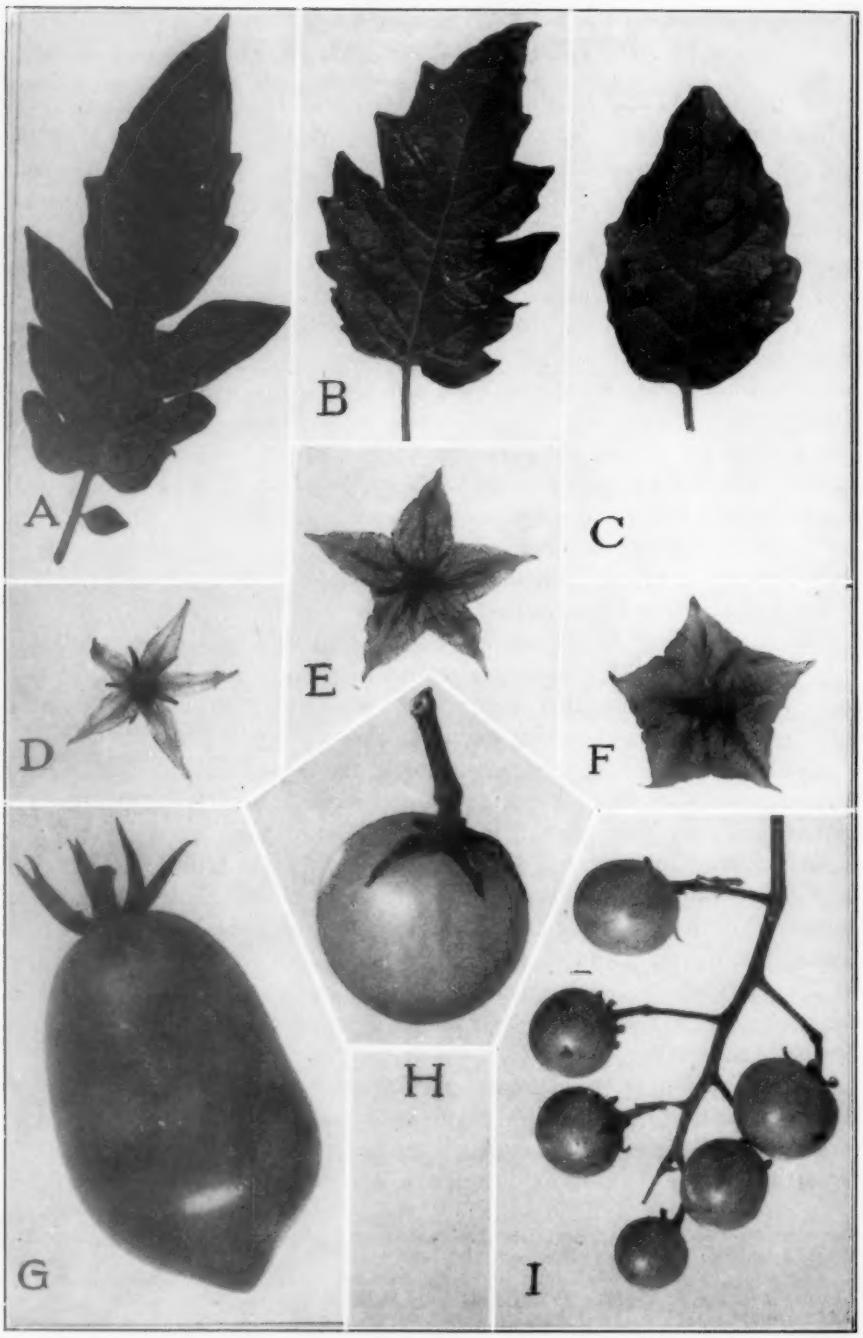

FIG. 1. Typical leaflets, flowers, and fruits of the F_1 hybrid, *Lycopersicon esculentum* ♀ × *L. peruvianum* ♂, and of its parents. Natural size. A, B, C. Leaflets of pistillate parent, of F_1 hybrid, and of staminate parent, respectively. D, E, F. Flowers of pistillate parent, of F_1 hybrid, and of staminate parent, respectively. G, H, I. Fruits of pistillate parent, of F_1 hybrid, and of staminate parent, respectively.

applied to the pedicels or peduncle when the flowers were pollinated (Fig. 1, B, E, H). However, all such fruits have been seedless. Plants propagated by cuttings when grown in an open field where many other tomato lines were growing, set fruit sparingly and very few fruits contained seed. These fruits ranged from ¾ to 1⅛ inches in diameter, and were uniformly globular, smooth, yellow, and usually 2-celled. A small population of F_2 plants has been grown from seeds obtained from the field-grown open-pollinated F_1 plants. These plants had marked phenotypic differences and most of them have been barren. Several outcrosses have been made to Pan America, Rutgers, and various hybrid combinations of *L. esculentum*. The progenies of all these outcrosses show wide segregations in plant habit, in foliage and fruit characters, and in fruitfulness. About 25 red-fruited and yellow-fruited selections have been made from these outcrosses. The selections are fruitful, the fruit ranging in diameter from 1½ to 2½ inches. Since these progenies were derived from open-pollinated field selections their genetic composition is somewhat uncertain because of the probability of some chance field crossing. However, this should not hinder the development of disease-resistant horticultural varieties from these interspecific outcrosses provided the resistance factors can be retained while breeding for fruit size, quality, and productiveness.—W. S. PORTE, Pathologist, and H. B. WALKER, Scientific Aid, Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration, U. S. Department of Agriculture.

Varietal Resistance of Tomato Seedlings to the Stem-Lesion Phase of Alternaria solani.[1]—One of the most serious disease problems connected with the production of tomato seedlings in the South for use in the northern canning areas is stem infection by *Alternaria solani.*[2] Considerable data[3, 4] have been compiled on methods which aid materially in reducing infection under field conditions but nothing has so far approximated complete control. In view of the many difficulties involved in control by cultural practices and spraying, resistant varieties appear to offer the best solution of this problem. Reynard and Andrus[5] showed that certain strains of tomato have considerable resistance to both the collar rot and the stem-canker caused by *A. solani,* and they suggested the possibility of breeding this resistance into canning varieties of tomatoes now being grown by northern farmers. In order to

[1] A phase of cooperative investigations between the Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration, U. S. Department of Agriculture; the Georgia Coastal Plain Experiment Station; the Georgia Agricultural Experiment Station; and the Georgia State Department of Entomology.

[2] Moore, W. D., H. Rex Thomas, and Edward K. Vaughn. Tomato seed treatment in relation to control of *Alternaria solani.* Phytopath. 33: 797–805. 1943.

[3] Moore, W. D. Some factors affecting the infection of tomato seedlings by *Alternaria solani.* Phytopath. 32: 399–403. 1942.

[4] Moore, W. D., and H. Rex Thomas. Some cultural practices that influence the development of *Alternaria solani* on tomato seedlings. Phytopath. 33: 1176–1184. 1943.

[5] Reynard, George B., and C. F. Andrus. Inheritance of resistance to the collar-rot phase of *Alternaria solani* on tomato. Phytopath. 35: 25–36. 1945.

study some of the resistant strains under commercial conditions, tests with a number of selections from the U. S. Regional Vegetable Breeding Laboratory, Charleston, S. C., were started in 1942 at Tifton, Ga., by the senior author and continued through 1944.

Seed beds were prepared during March of each year by turning the soil, harrowing, and broadcasting a 4–8–4 fertilizer at the rate of 1,000 pounds per acre. The soil was smoothed with a drag and the tomato seed planted approximately an inch deep by hand in rows 16 inches apart and at the rate of 20 seed per foot. The beds were watered as needed with an overhead sprinkling system and the rows cultivated with a hand cultivator to keep down grass and weeds. About mid-May, when the seedlings were approximately 8 inches tall, 25-plant lots were pulled at random from each plot. The roots and lower portion of the stems were wrapped with wet moss and paper and the plants were then stored at room temperature for 48 hours. Following storage, 4 replicates of 25 plants from each strain (15 plants were used in 1944) were set twelve inches apart in rows 3 feet apart. The plants were allowed to grow for periods ranging from 21 to 36 days, after which they were carefully pulled from the soil, thoroughly washed, and the number of stem lesions recorded.

The 1942 test consisted of 13 tomato strains as follows: one each of Marglobe, Indiana Baltimore, King George, Danish Early, Targinnie Red, and Devon Surprise; two selections of Norduke, two of Marglobe × Red Currant cross, and three of Riverside. The number of stem lesions varied from a mean of 158.5 per replicate on Marglobe to 4.5 on King George. There were five strains with means varying from 4.5 to 8.2, four with means from 10.2 to 22.0, and four with means from 64.7 to 158.5.

As a result of growth studies made during the summer of 1942 several of the strains were dropped and others were added to give the following selections for the 1943 test: three from Targinnie Red and one each from a Red Currant × Marglobe cross, a Devon Surprise × Marglobe cross, a Targinnie Red × Montgomery cross, Devon Surprise, Norduke, and Marglobe. The mean number of stem lesions per replicate in this test varied from 213.7 on Marglobe to 0.2 on Norduke. Four strains had lesions varying from 0.2 to 1.0, four had from 5.5 to 24.0 lesions, and one had 213.7 lesions.

Additional eliminations were made during the summer of 1943 and the 1944 test included only 5 selections. They were as follows: three F_3 selections from the original Marglobe × Devon Surprise cross outcrossed to Pan America, one F_4 selection from a Cooper Special × Devon Surprise cross, and one Marglobe. The mean number of stem lesions in this group varied from 248.2 on Marglobe to 25.5 on the Cooper Special × Devon Surprise cross. The three selections from the Marglobe × Devon Surprise × Pan America strain had lesions varying from 34.0 to 38.0.

While these strains do not have complete resistance to stem infection by *Alternaria solani* and are not comparable to the average canning tomatoes in size and quality of fruit, they possess definite possibilities as stock that

may be used for crossing with other varieties in a disease-resistance breeding program. The pure lines of Targinnie Red, Devon Surprise, Norduke, and their several crosses were significantly more resistant to stem infection by *Alternaria solani* than Marglobe in all tests during the three seasons in which this study was made.—W. D. MOORE, Pathologist, and GEORGE B. REYNARD, formerly Assistant Geneticist, Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration, U. S. Department of Agriculture, Beltsville, Maryland.

Meadow Nematodes as the Cause of Root Destruction.—The year 1889 is notable in the history of research on plant-parasitic nematodes in the United States as the year of publication of the first papers on this subject matter in this country, namely Atkinson's bulletin on the root-knot nematode[1] and also that of Neal.[2] There is, however, a third publication of the same year which is generally overlooked: F. Lamson Scribner's "Diseases of the Irish Potato,"[3] an important part of which deals with nematode diseases. Scribner also reports root knot, but in addition a second nematode affliction of potatoes is described. The causative nematode is not named but it is easily recognized by description, figures, and symptoms as a meadow nematode, which has been named Scribner's meadow nematode.[4]

While the root-knot nematode has received considerable attention as a disease factor, the meadow nematodes—plant-parasitic nematodes of the genus *Pratylenchus* Filipjev—have been almost completely ignored. This is unfortunate. The pratylenchs are doubtless one of the most important primary factors in root destruction among cultivated and uncultivated plants. The overall damage caused by these pests in the United States is possibly greater than that which is attributable to the root-knot nematode. There are several obvious reasons for this underestimation. Meadow nematodes do not produce specific symptoms. The lesions they cause on roots are not different from those produced by many other nematodes or from those induced by many fungi and bacteria. Excised root sections placed on culture media will not bring them to light since they will perish under these abnormal conditions. Meadow nematodes are migratory, often almost completely evacuate a decaying root, and may not be found at all even if the root is carefully examined. They are very small, mostly 0.350 to 0.800 mm. in total length. They may enter a root at any place, even in very woody portions. However, they appear not to enter the vascular system but to

[1] Atkinson, George F. A preliminary report on the life history and metamorphoses of a root-gall nematode, *Heterodera radicicola* (Greeff) Müll., and the injuries caused by it upon the roots of various plants. Science Contrib., Alabama Agricultural Experiment Station 1: 177–226. 1889. Also published as Ala. Agr. Exp. Stat. Bull. 9. 1889.

[2] Neal, J. C. The root-knot disease of the peach, orange, and other plants in Florida, due to the work of *Anguillula*. U. S. Dept. Agr., Div. Entom. Bull. 20. 1889.

[3] Scribner, F. L. Diseases of the Irish potato. Tenn. Agr. Exp. Stat. Bull. 2(2): 1–43. 1889.

[4] Sherbakoff, C. D., and W. W. Stanley. The more important diseases and insect pests of crops in Tennessee. Tenn. Agr. Exp. Stat. Bull. 186. 1943.

migrate mainly in the cortical tissues, rarely in the pith and its rays. Other subterraneous formations of a plant (bulbs, corms, tubers, rhizomes, in rare instances even basal portions of stems) appear subject to attack. Eggs may be deposited in the tissues of a host plant or in the soil. Not infrequently aggregations of specimens form nests of infection which may ring a root and thus amputate its distal portions. It is obvious that even a small number of specimens may, by this mode of attack, greatly interfere with the functioning and growth of a root system. The plant often attempts to repair the damage

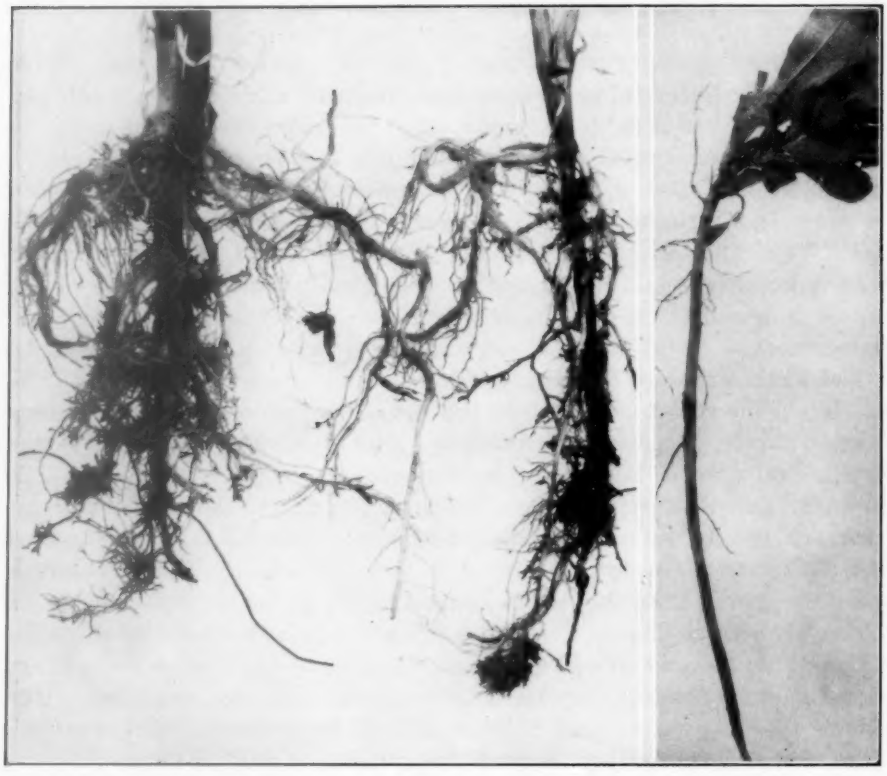

FIG. 1. At left, bearded root systems of two plants of corn, *Zea mays* L. (var. Hill's Yellow Dent) infected with numerous specimens of two different species of meadow nematodes (*Pratylenchus sp.*) and also a few specimens of the root-knot nematode (*Heterodera marioni* [Cornu] Goodey). Plants collected near Jacksonville, Texas, and submitted by P. A. Young. At right, a root of the goldenrod (*Solidago leavenworthii* T. & G.) with black lesions caused by a meadow nematode; specimen collected and submitted by H. G. Ukkelberg at Fort Myers, Fla.

by forming secondary roots and rootlets above the nematode lesions; a matted and bearded root system with only short feeder roots will result, *i.e.*, a root system that exploits only the soil near the surface and close to the root crown, while deeper soil strata and more distant surface soils remain out of reach of the plant (Fig. 1). This condition shows on the above-ground parts of the affected plant in reduced growth, in wilting during the hot part of the day, in dying twigs or branches, in bronzing of leaves (evergreens) during

the winter, in winter kill, and in drought damage during the summer or what is sometimes called dieback or sunstroke. Various root crops are also liable to disfiguration by these meadow nematodes, particularly potatoes, peanuts (Fig. 2), and lily bulbs, in which instances the market value may be decreased or the product made directly unsalable.

The direct damage caused by these pests on roots is, however, only part of the story; all root-invading nematodes open the path for other, sometimes serious, secondary invaders which may result in necrosis of adjacent tissues and preclude recovery.

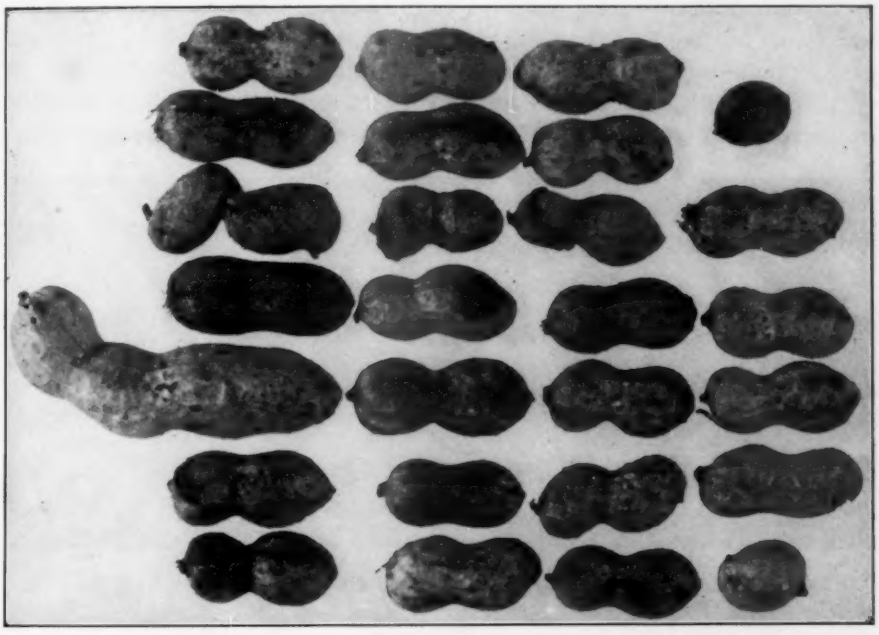

FIG. 2. Peanuts (*Arachis hypogaea* L.) disfigured by numerous black spots caused by a species of meadow nematode (*Pratylenchus sp.*); collected at the Substation at Holland, Va., of the Virginia Agricultural Experiment Station and submitted by D. J. Humphrey and L. J. Cushman.

The location and identification of pratylenchs in a root system or other plant organs necessitate a careful teasing apart of the tissues under a binocular microscope. High magnifications are necessary and identification of the species is difficult. Although the group as such is well characterized and appears to be a very natural one, its taxonomy has not yet been worked out. Nor is much known about the life cycle, host range, and geographical distribution of the various species of which at least five occur in the United States. There is need for more work on this group because of its economic significance.—G. STEINER.

A Bacterial Streak Disease of Phleum pratense L.[1]—A bacterial streak disease of timothy was first collected near Platteville, Wisconsin, in 1925.

[1] Journal Paper No. J-1283 of the Iowa Agricultural Experiment Station, Ames, Iowa, Project No. 450.

Later, in 1940, diseased timothy was collected in Mitchell County in the vicinity of Osage, Iowa, and near Kanawha, in Wright County, Iowa, along roadsides, in hay fields, and in pastures. In 1941 the disease was common in Story County around Ames, Iowa. It is difficult to estimate the loss to the timothy hay and forage crops caused by this disease because its prevalence fluctuates from year to year and is greatest during wet seasons.

The streaks of the disease varied from barely visible ones to those over two centimeters long on the leaf blades of the young shoots. A blotching of the whole leaf was common on the plants artificially inoculated in the greenhouse but was rarely observed in the field. When the necrosis of the leaf tissue became prevalent the plants were stunted. A striking symptom of the plants in the hay stage was the streaking on the blades and sheaths of infected flag leaves. Under severe attack the emerging heads were sealed in the spiral whorl by bacterial exudate or were malformed upon emergence

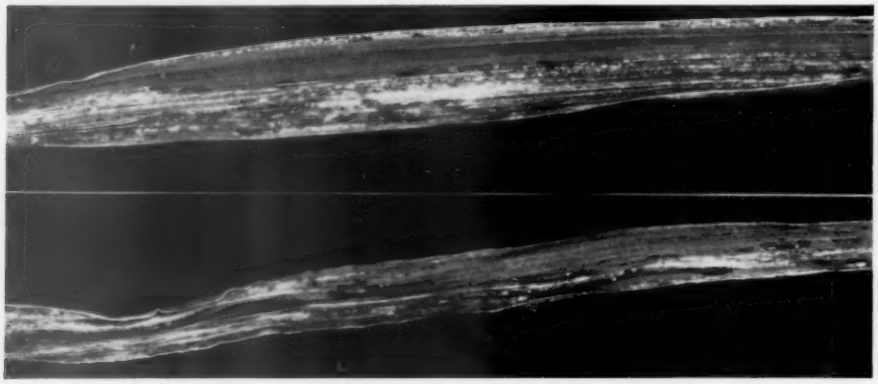

FIG. 1. Water-soaked, translucent streaks on timothy leaves artificially inoculated with the timothy strain of *Xanthomonas translucens*.

from the boot. During warm humid days, yellowish droplets of bacterial exudate formed on the surface of the lesions even as late as October and November. Upon drying, these droplets formed hard, resinous granules. In the later stages of the disease the affected tissues became dry and brown, but even these older brown lesions were translucent by transmitted light. The symptoms of this disease were very similar to those caused by *Xanthomonas translucens* on barley, brome grass, rye, and wheat.

A yellow bacterium was isolated from the translucent, water-soaked streaks and from the dry, brown lesions. The bacterium was a Gram-negative, monotrichous rod averaging 0.5 to 0.8 μ by 1.5 to 2.0 μ, depending upon the age of the culture studied. In culture the organism was a yellow waxy color. Colonies on nutrient agar were raised, with a smooth glistening surface. After positive proof of pathogenicity, the comparative cultural characteristics of three isolates of the causal agent were studied. All grew slowly in beef-peptone agar, while on potato-dextrose agar growth was copious and a watery, creamy yellow with whitish margins. In nutrient

broth the organism formed a coarse pellicle. The organism liquified gelatin, but did not reduce nitrates, produced hydrogen sulphide, produced ammonia, reduced the indicator and produced an alkaline reaction in litmus milk, did not produce acetyl-methyl-carbinol, was methyl-red-negative, did not utilize citrate, showed diastase activity, utilized only dextrose of the seven monosaccharides tested in inorganic basal medium and utilized the disaccharides maltose and sucrose. Culturally and biochemically the organism was like *Xanthomonas translucens* although there was some variation between isolates of the timothy strain and other strains of *X. translucens* in utilization of carbohydrates.

After identifying the bacterium, repeated cross inoculations were made to determine whether or not the bacterium was a new variety of *Xanthomonas translucens.* The organism caused typical symptoms on timothy (Fig. 1) by both the spray and hypodermic methods of inoculation, but it did not infect barley, brome grass, oats, rye, or wheat. As a result of these trials the bacterium causing a bacterial streak disease on timothy is designated a variety of *Xanthomonas translucens* as follows: *Xanthomonas translucens* var. **phleipratensis** var. nov. Isolated from *Phleum pratense.* Produces water-soaked lesions at 25° to 30° C. on shoots of *Phleum pratense* following wound inoculation.—J. R. WALLIN and C. S. REDDY, Iowa Agricultural Experiment Station, Ames, Iowa.

Notes on Physiologic Specialization in Puccinia graminis tritici Erikss. and Henn. in China.[1]—In 1932, Tu[2] reported the occurrence of 6 physiologic races of *Puccinia graminis tritici* in China. As far as the writer is aware, it is the only published record regarding physiologic specialization of wheat stem rust in this country.

During 1942–1944, the writer made 175 collections of *Puccinia graminis tritici* from 12 provinces in China. Inoculations on the 12 standard wheat varieties,[3] seed of which was obtained through the courtesy of Dr. E. C. Stakman of the U. S. Department of Agriculture and the University of Minnesota, U.S.A., demonstrated 2 new physiologic races and 12 already known races, *viz.,* Nos. 10, 11, 15, 34, 39, 40, 95, 107, 115, 122, 143, and 189.[4] Races 15, 107, and 122 are by far the most common: they have been identified 35, 44, and 24 times, respectively, out of 175 collections. Race 122 is the most widely distributed, occurring in 9 of 12 provinces.

The two new races are tentatively numbered C_1 and C_2. The reactions

[1] PHYTOPATHOLOGY extends the courtesy of its journal pages to scientists in other countries who are persevering in research under difficult wartime conditions and are temporarily deprived of the opportunity for membership in the American Phytopathological Society.

The writer is indebted to Dr. E. C. Stakman for reading the manuscript.

[2] Tu, Chih. Physiologic forms of *Puccinia graminis tritici* in Kwangtung, Southern China. Phytopath. 24: 423–424. 1934.

[3] Stakman, E. C., and M. N. Levine. The determination of biologic forms of *Puccinia graminis* on *Triticum* spp. Minn. Agr. Exp. Stat. Tech. Bull. 8. 1922. (Mimeographed keys and tables for identifying races are available.)

[4] Garcia-Rada, G., J. Vallega, W. Q. Loegering, and E. C. Stakman. An unusually virulent race of wheat stem rust No. 189. Phytopath. 32: 720–726. 1942.

of the standard wheat varieties to these races were tested 5 times at temperatures ranging from 22° to 28° C., and the results obtained were consistent (Table 1).

TABLE 1.—*The infection types produced on 12 standard differential wheat varieties by races C_1 and C_2 of Puccinia graminis tritici*

Acc. No.	Place of collection	Infection types on wheat hosts[a]											
		LC	Ma	Krd	Ko	Arn	Mnd	Spm	Kub	Ae	Enk	Ver	Kpl
C_1	Kaiyüan, Yunnan Kunming, Yunnan	4++	2++	4++	0;	4++	4++	4++	4++	4++	3+	1++	3+c
C_2	Hwayang, Szechuan Chengtu, Szechuan	4+	3±c	4+	0;	4++	4+	4+	4++	4+	4–	3	1+

[a] The wheat hosts are those used by Stakman and Levine to determine physiologic races of wheat stem rust.

Race C_1 was first isolated from common wheat in Kaiyüan and then again from Khapli emmer in the nursery. It can be differentiated from known races by the infection types it produces on Kota and on Vernal. It is similar to races 41 and 42 of Stakman and Levine except on Kanred, on which this new race produces a 4++ infection type instead of the 0 infection type. Race C_1 also differs from races 72 and 99 by its effects on Little Club and Arnautka. Race C_2 is similar to race 122 except that the former produces a type 3 infection and the latter a type 1 infection on Vernal.—SIN-YÜN YIN, Division of Plant Pathology, Institute of Agricultural Research, National Tsing Hua University, Kunming, Yunnan, China.

A Graft-Transmissible Mosaic Disease of Grapevine.—A mosaic type of disease reported to be killing grapevines (*Vitis vinifera*) in a vineyard in Napa Valley, California, was brought to the writer's attention in the summer of 1943. The disease involved a somewhat circular area of about 60 vines near the center of a block of the variety Palamino grafted on St. George root stock.

The leaves of diseased vines from emergence to leaf fall had varying degrees and patterns of chlorosis that consisted essentially of yellow, cream, and light green areas. The cream chlorosis occurred as a narrow band along the smaller veins (Fig. 1, A), as irregular blotches along the large veins (Fig. 1, B and D), or as a stippling or spattering over the leaf surface (Fig. 1, E). Some leaves were entirely yellow with only traces of green along the large veins. In other leaves (Fig. 1, F) the light green chlorosis appeared to have originated in the veins and spread, as if leaking from the veins, into the surrounding tissue. Many leaves had two or more of the types of mottling with one type usually predominating. An occasional leaf appeared to be covered with a mixture of types and had some veinlet clearing as well (Fig. 1, C). The cream and yellow mottled areas were usually

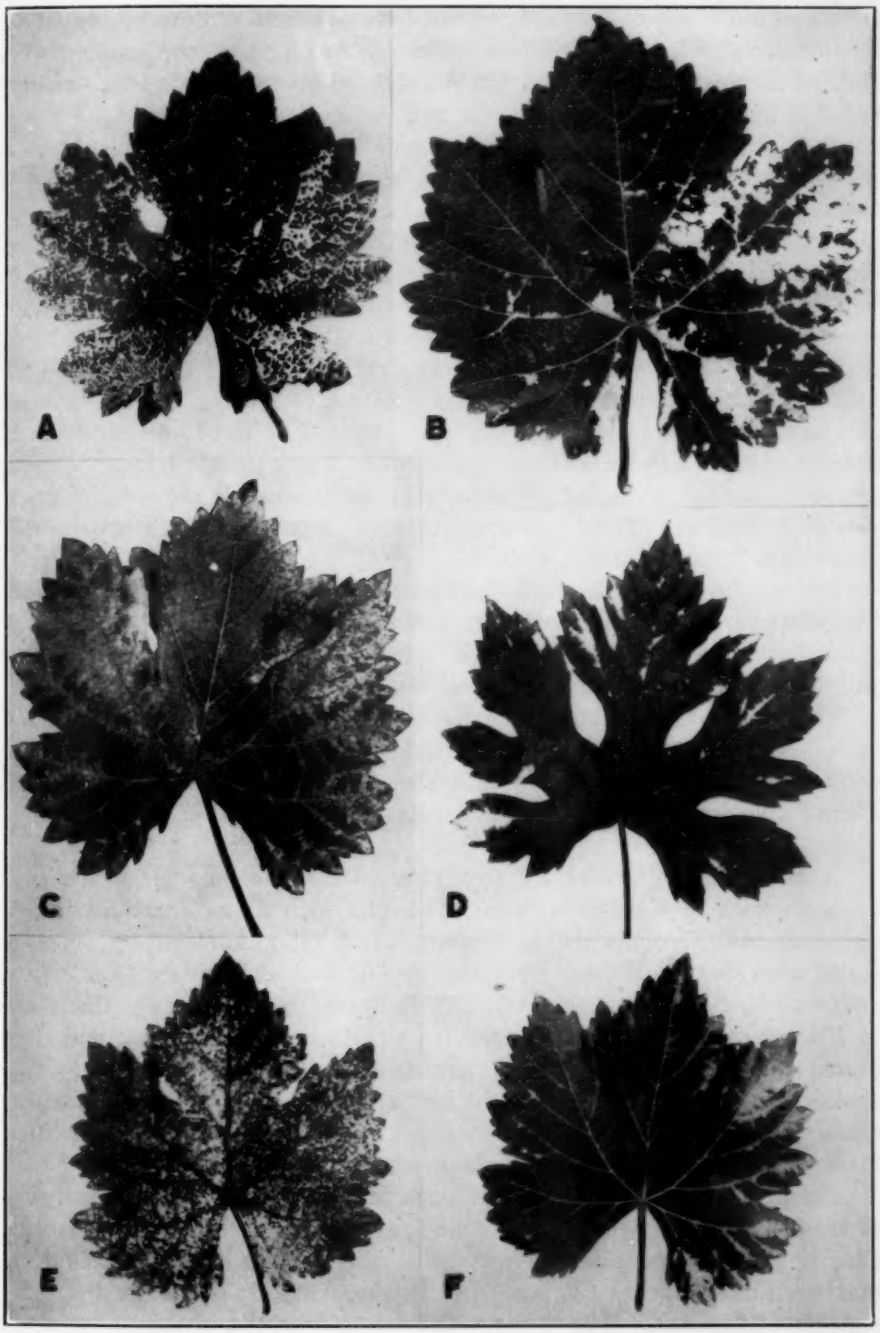

FIG. 1. Leaves of *Vitis vinifera* with different patterns of mottling associated with a graft-transmissible mosaic. A. Leaf with cream-colored bands along the small veins and also some necrosis of the basal lobes. B. Leaf with irregular creamy blotches, mostly along the veins. C. Leaf with a combination of cream, yellow, and light green mottling and also some veinlet clearing. D. Leaf from a seedling vine inoculated by grafting a bud from a diseased vine into the stem. The leaf has cream-yellow patches along the large veins. E. Leaf in which the yellow color appears to have been stippled over the surface. F. Leaf with light green areas along the veins.

faint and indistinct in the young leaves, but as the leaves matured the areas became prominent, with definite margins contrasting with the green of the leaf. During the summer and fall the mottled areas in the leaves usually faded to light cream or nearly white, and parts of the leaves dried.

In September, 1943, twenty-five cuttings were taken from diseased vines in the Napa Valley vineyard and rooted in sterile soil. Sixteen of the cuttings grew and all of these had mottled leaves. At the same time, buds were also taken from diseased vines and grafted, one each, into 5 healthy seedling vines growing in greenhouse pots. By January, 1944, the stock wood of 3 budded vines had developed mottled leaves typical of those of the diseased vines from which the buds were originally taken. Again, in 1944, buds from diseased vines were grafted into 10 additional healthy seedlings and 15 healthy rooted cuttings of the variety Palamino. During the fourth and fifth months of incubation, the stock growth in 7 of the 10 seedling vines and 12 of the 15 Palamino rootings had developed mosaic leaves. Check plants of seedling vines and Palamino rootings handled in the same manner remained healthy. These experiments show that this mosaic disease of grapevines is graft-transmissible.

During July, 1944, the carborundum method described by Rawlins and Tompkins,[1] was used to inoculate 25 separate seedling vines with juice extracted from chlorotic leaves. After 8 months of incubation, none of the juice-inoculated plants had developed leaf-mottling symptoms.

This graft-transmissible mosaic disease of grapevines found in California resembles very closely the juice-transmissible mosaic of *Vitis vinifera* in central Europe described by Strănák *et al.*[2]—WM. B. HEWITT, Division of Plant Pathology, University of California, Davis, California.

A More Virulent Black Pit Organism on Citrus.—In some lemon and orange groves of California large spots (1.5 inch diameter) were found on lemon and Valencia orange fruits (Fig. 1, C) either solitary or associated with the typical black pit caused by *Phytomonas syringae* (*Bacterium citriputeale, Bact. citrarefaciens*). These spots, observed for the first time in 1945, averaged much larger than the usual black pit of lemon and suggested either a greatly increased virulence or a different organism. The tissues were depressed, brown, and had a sharply defined darker margin. Some of these spots were evidently caused by the coalescence of the smaller black pit lesions, while in other spots no coalescence could be observed.

Cultures made from the typical black pit lesions and from the margins of the larger spots gave bacterial growth typical of the black pit organism. On inoculation, these cultures caused the development of spots reaching a maximum of 1.5 inches (35 mm.) in diameter (Fig. 1, B) rather than the usual small black pits (Fig. 1, A). The maturity of the fruit and the viru-

[1] Rawlins, T. E., and C. M. Tompkins. Studies on the effect of carborundum as an abrasive in plant virus inoculations. Phytopath. 26: 578–587. 1936.

[2] Strănák, F., Ctibor Blattný, and A. Klečka. Mosaika révy vinné. Ochrana Rostlin. 11: 89–98. 1931.

lence of the organisms are believed to determine the size of spots. Since spots of this size had not previously been found in nature, the recent isolates appear especially virulent. Smith[1] in his description of the black pit of

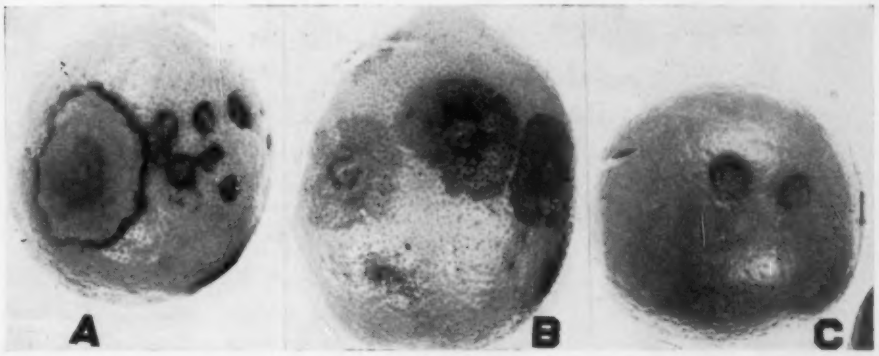

FIG. 1. Black pit on lemon: A, Natural infection showing the large spot with the darker margin and small typical black pits; B, Artificial inoculation showing the large spots before the tissue has become depressed; C, Natural infection on Valencia orange.

lemons gave the size of the lesions as 5–20 mm., with larger lesions rarely occurring.—CLAYTON O. SMITH and L. J. KLOTZ, University of California Citrus Experiment Station, Riverside, California.

[1] Smith, Clayton O. Black pit of lemons. Phytopath. 3: 277–281. 1913.

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